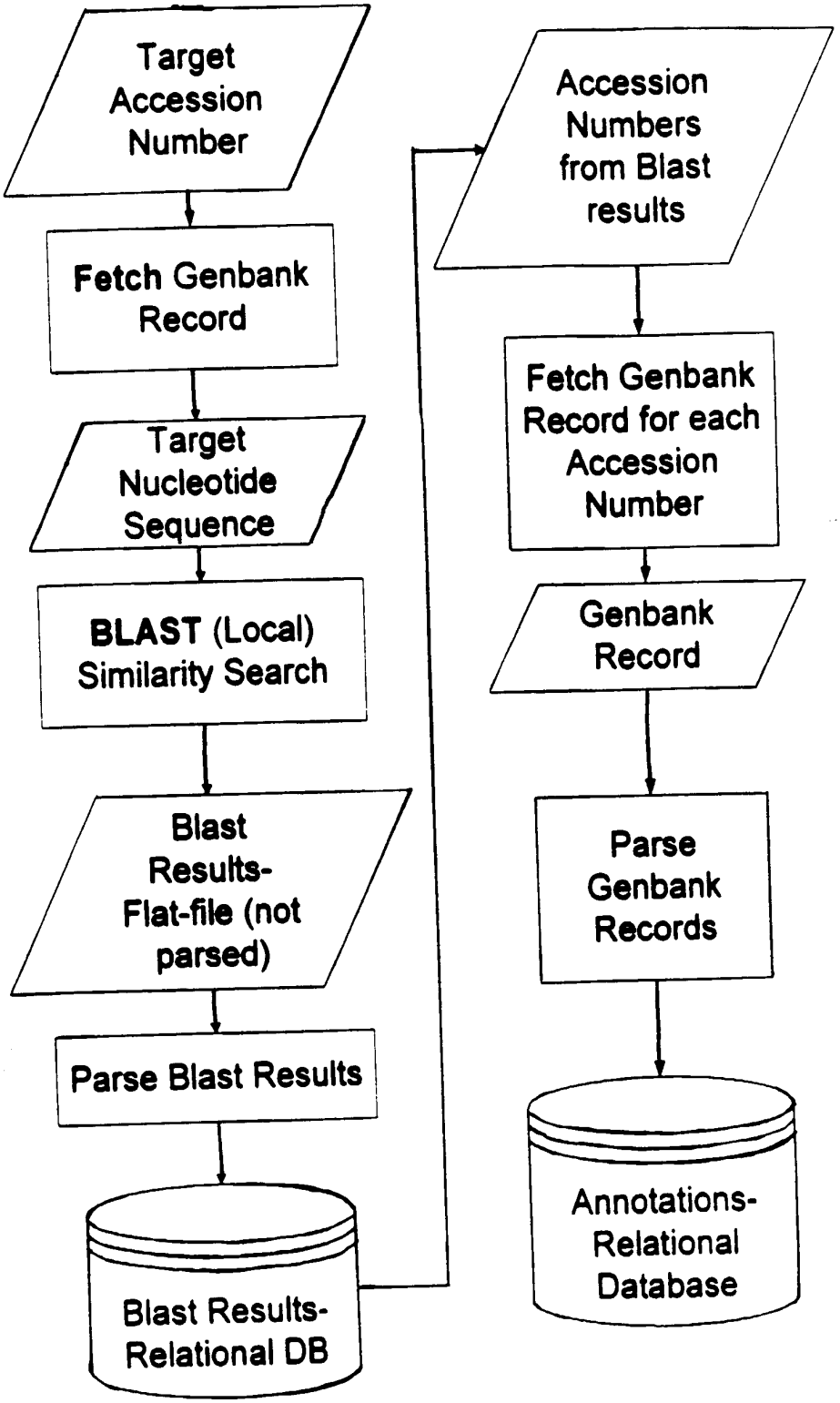

Target Accession Number
Fetch Genbank Record
Target Nucleotide Sequence
BLAST (Local) Similarity Search
Blast Results- Flat-file (not parsed)
Parse Blast Results
Blast Results- Relational DB
Accession Numbers from Blast results
Fetch Genbank Record for each Accession Number
Genbank Record
Parse Genbank Records
Annotations- Relational Database

Figure 3

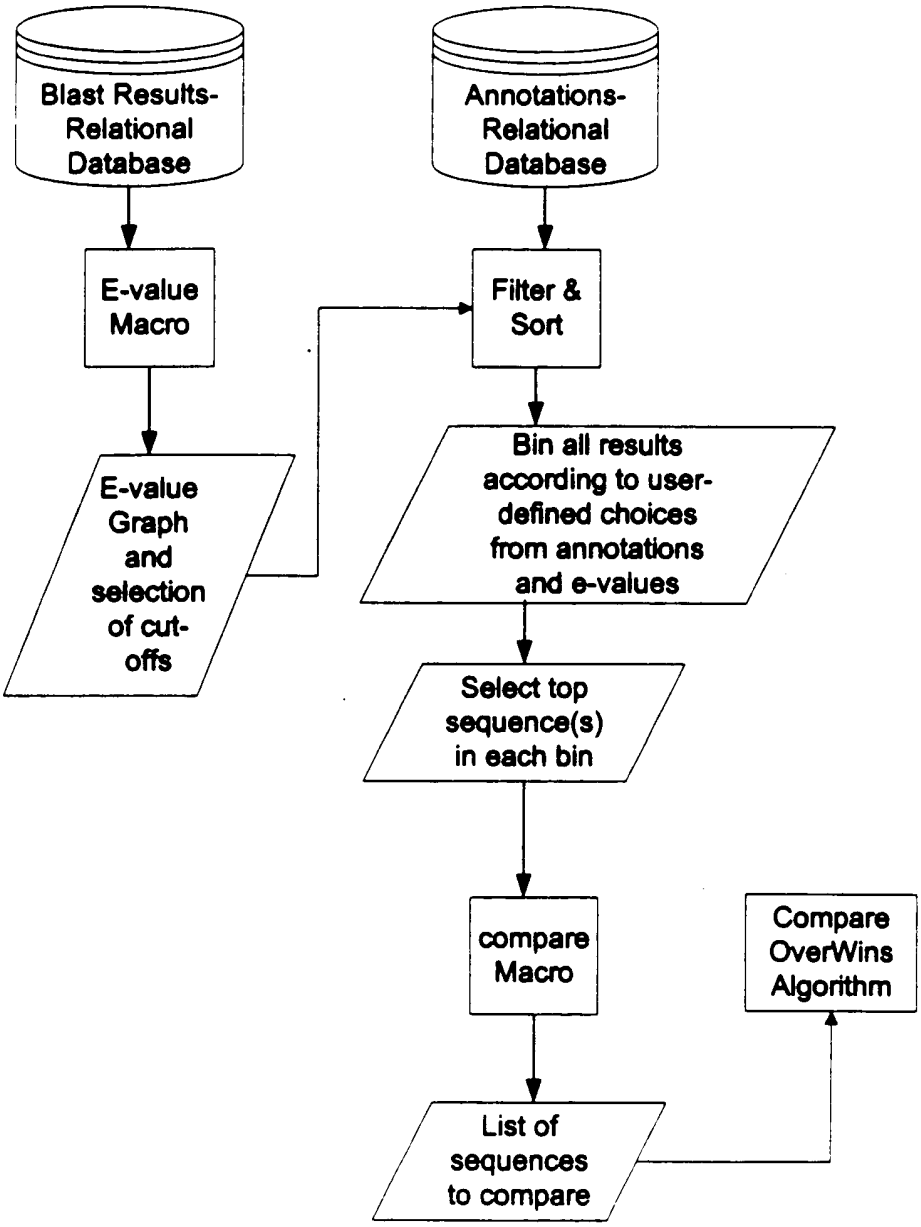

Blast Results-Relational Database
Annotations-Relational Database
E-value Macro
Filter & Sort
E-value Graph and selection of cut-offs
Bin all results according to user-defined choices from annotations and e-values
Select top sequence(s) in each bin
compare Macro
Compare OverWins Algorithm
List of sequences to compare

FIGURE 4

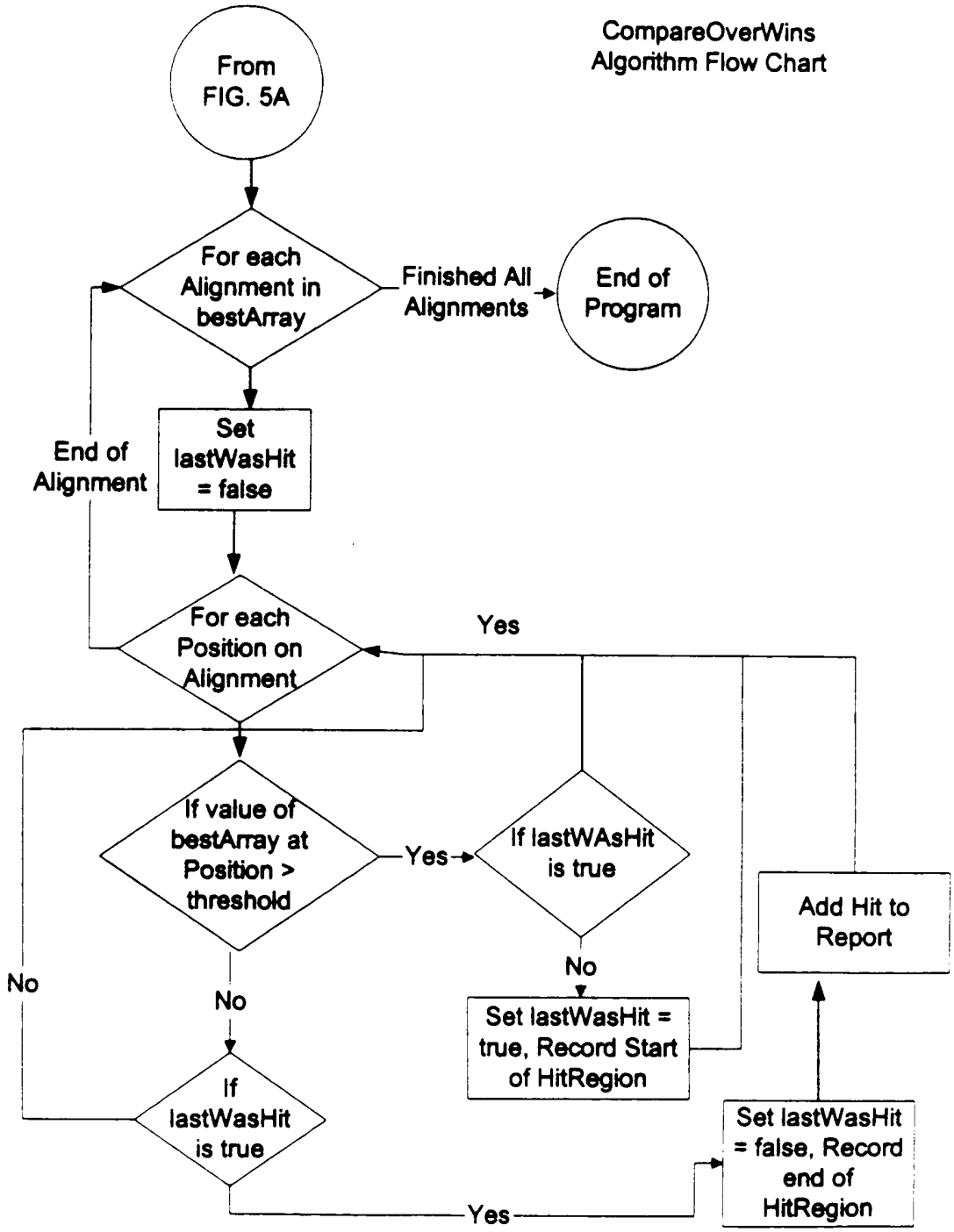

CompareOverWins
Algorithm Flow Chart
From FIG. 5A
For each Alignment in bestArray
Finished All Alignments
End of Program
End of Alignment
Set lastWasHit = false
For each Position on Alignment
Yes
If value of bestArray at Position > threshold
Yes
If lastWAsHit is true
No
Add Hit to Report
No
No
Set lastWasHit = true, Record Start of HitRegion
If lastWasHit is true
Set lastWasHit = false, Record end of HitRegion
Yes

FIGURE 5B

Input:
Sequence A length a
Sequence B length b
Window Size

CompareOverWins
Algorithm Flow Chart
Basic Compare

Output:
Array of size a by b of unsigned chars (0-255)
Each point represents the number of matches in the window at that allignment and position

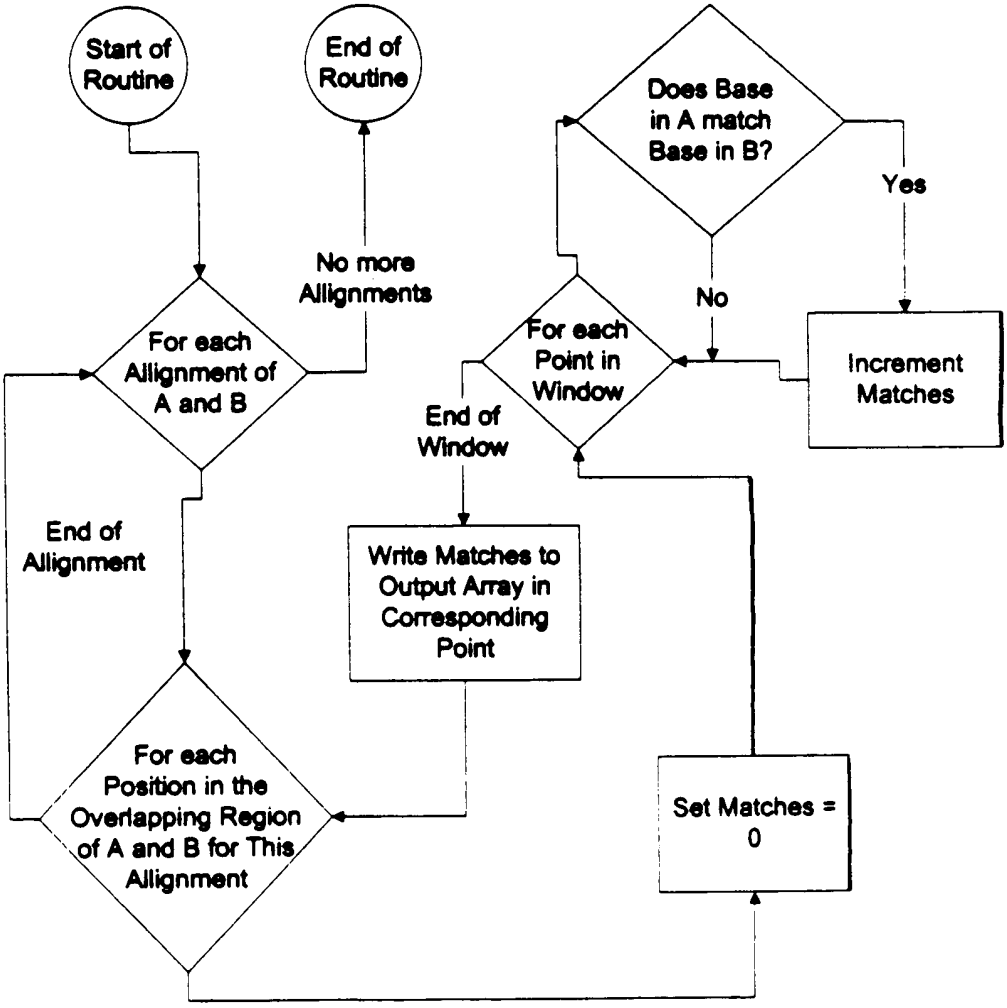


FIGURE 5C

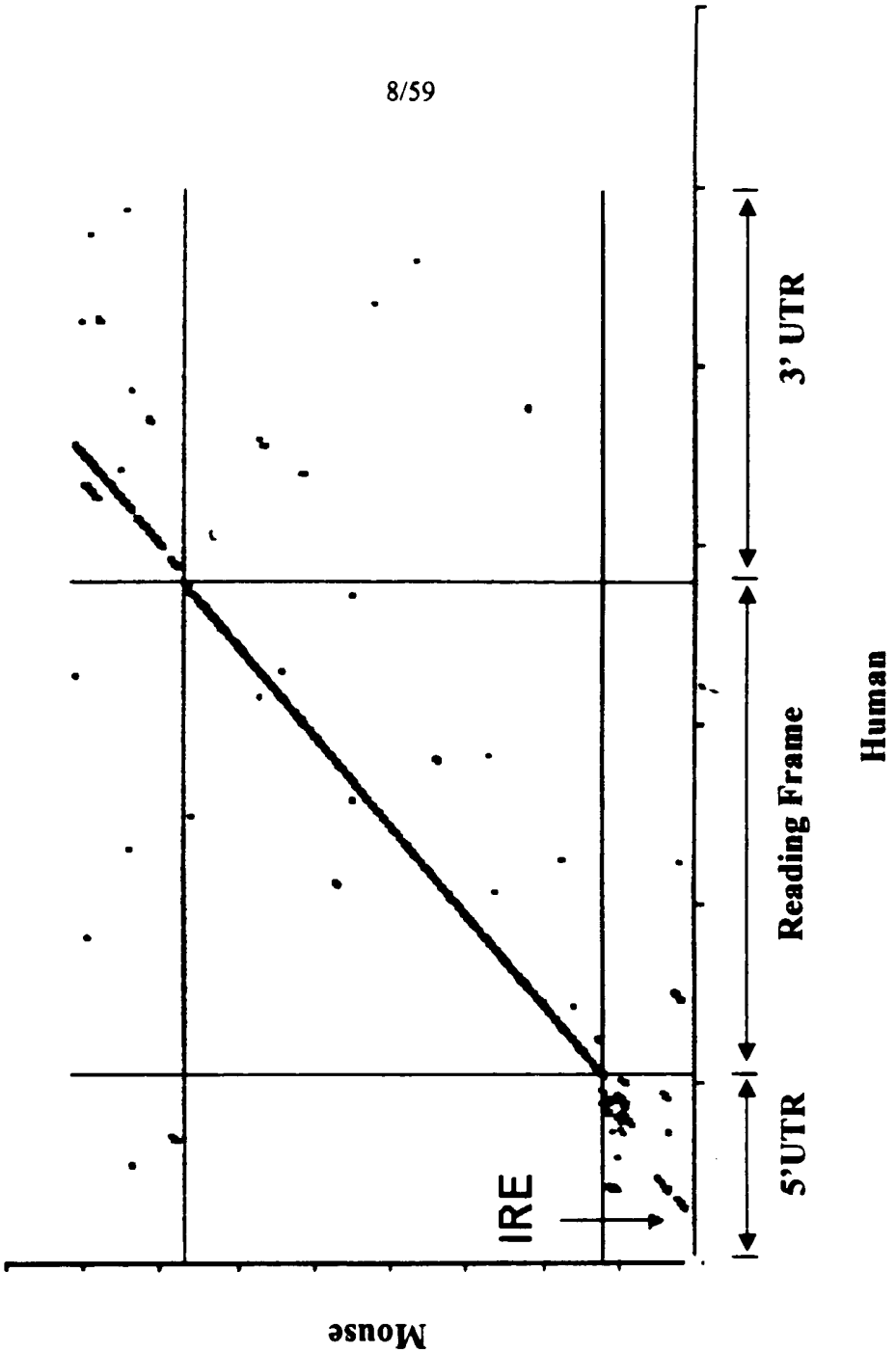


Figure 6

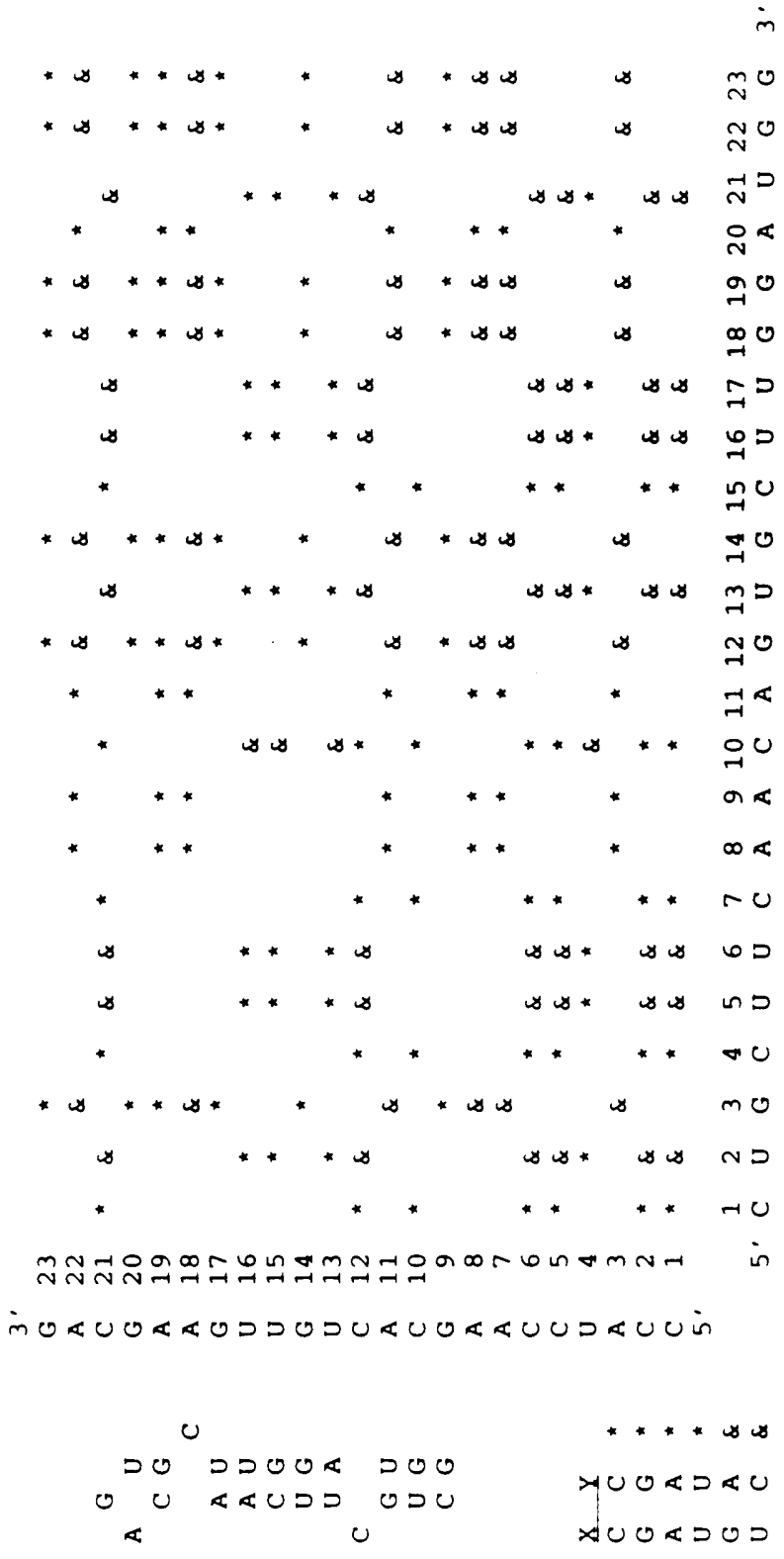

Self Complementation Analysis - Single Sequence
3' G A C G A A C C C U 5'
5' C U G C U U C A A C A G U G C U U G G A 3'
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23
X Y
C C G A A U U G A
C G A A U U C C U
G A C G C G U A C
C U G C G U A U G
A C G U A C G U A
G U C G U A U C G
A C G C

Figure 7

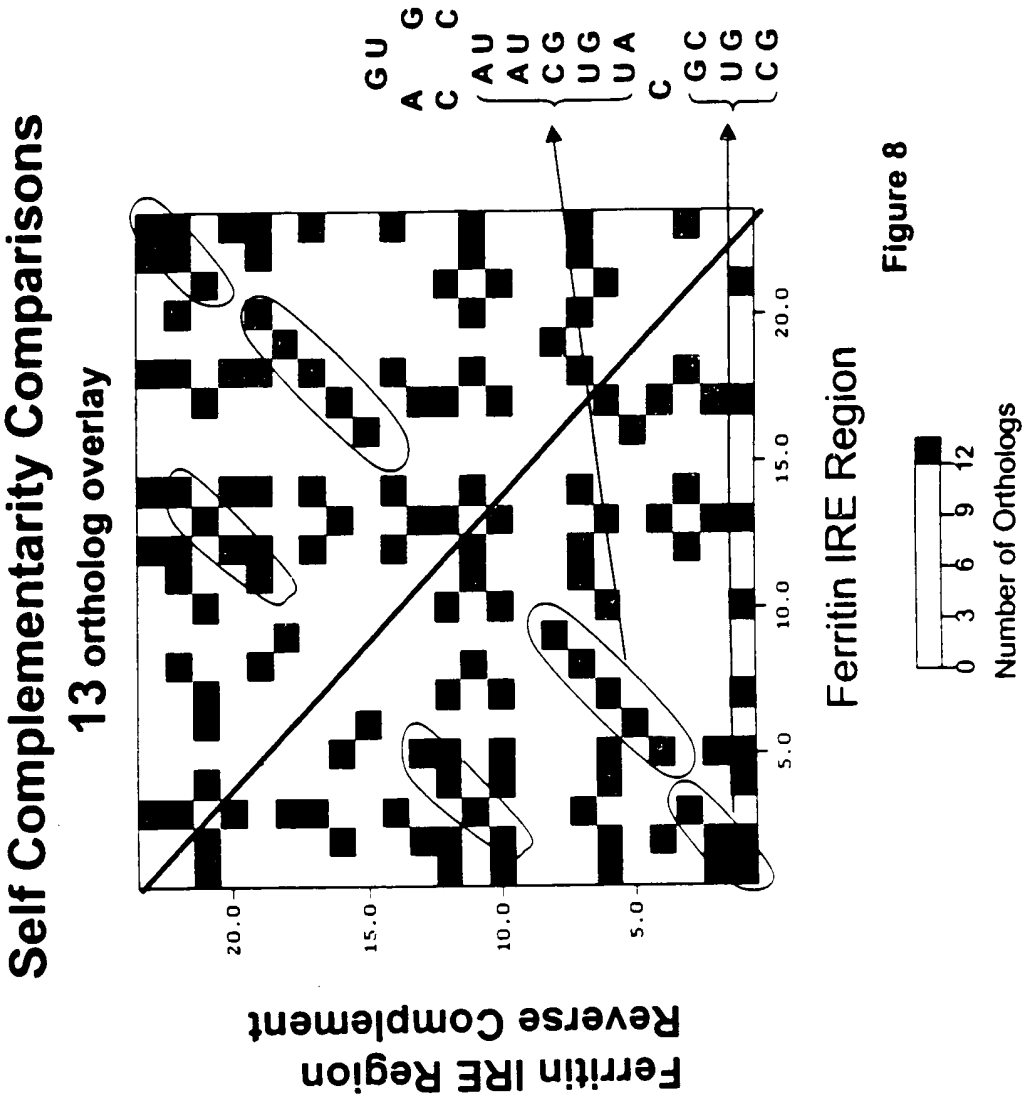
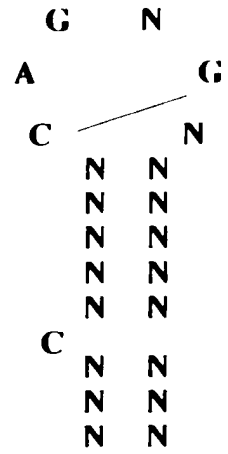

Self Complementarity Comparisons
13 ortholog overlay
Ferritin IRE Region
Reverse Complement
Ferritin IRE Region
Number of Orthologs
0 3 6 9 12
5.0
10.0
15.0
20.0
G U
A C
C G
A U
A U
C G
U G
U A
C
G C
U G
C G

Figure 8

Figure 9



IRE
Stem-loop Model

```
H1 S1 H2 S2 H2 H1

H1  3:3 NNN:NNN
S1  1 C
H2  5:5 NNNNN:NNNNN
S2  6 CAGNGN

W2
M0
```

IRE String descriptor

This descriptor allows for
- a wobble (W) of 2
- no mismatches.
- N can be any nucleotide
- H refers to the stem region
- S refers to the single stranded region.

Docket No.: IBIS-0007
Title: MODULATION OF MOLECULAR INTERACTION SITES ON RNA AND OTHER BIOMOLECULES
Inventor(s): Ecker et al.
Attorney: Paul K. Legaard 215-568-3100
Sheet 11 of 59

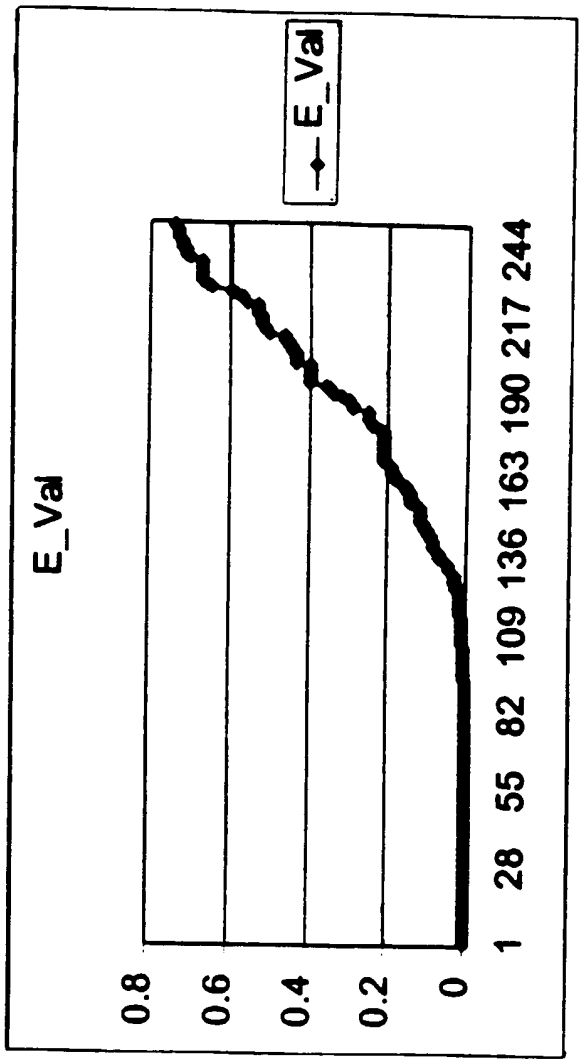
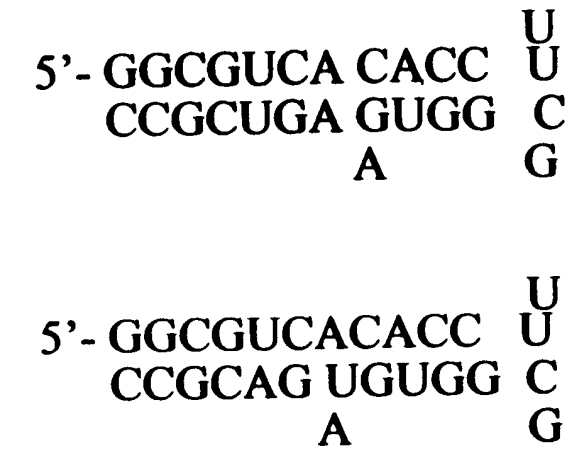
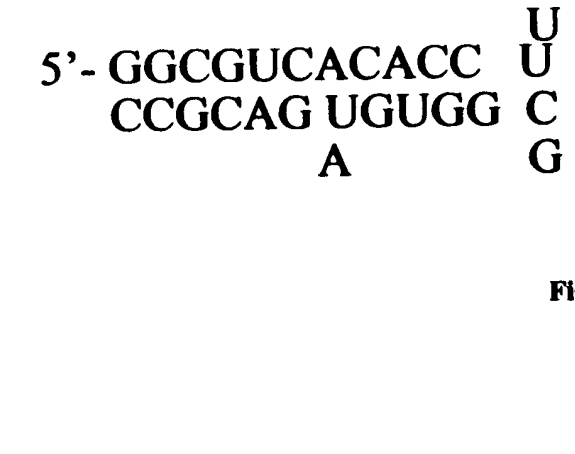


Figure 10



16S A Site rRNA



Control RNA

Figure 40

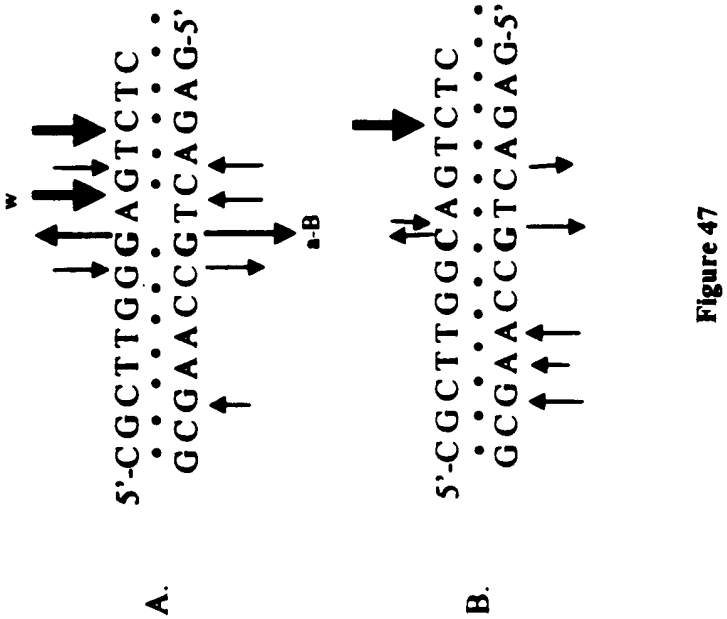

A.
5'-CGCTTGGGAGTCTC
GCGAACCCGTCAGAG-5'
w
a-B
B.
5'-CGCTTGGCAGTCTC
GCGAACCCGTCAGAG-5'

Figure 47

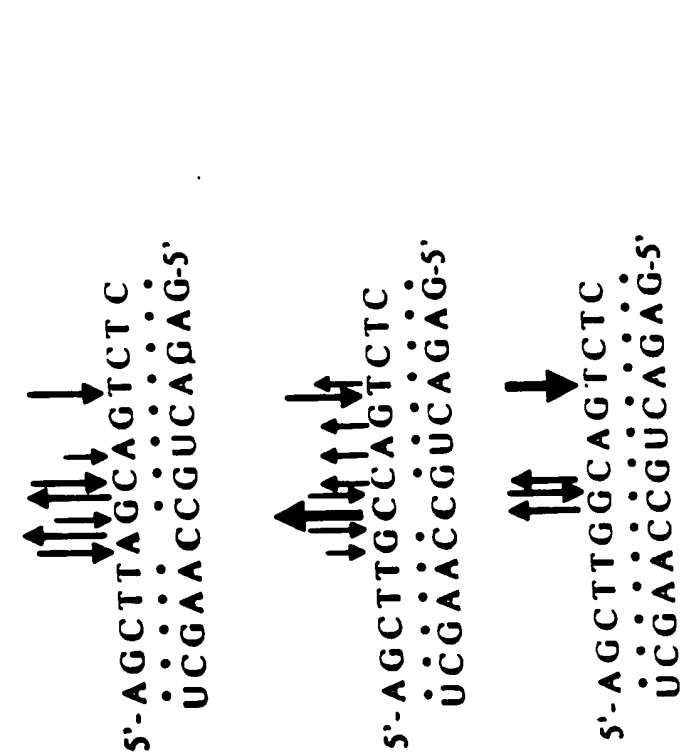


Figure 48 MS Fragmentation of DNA:RNA duplexes

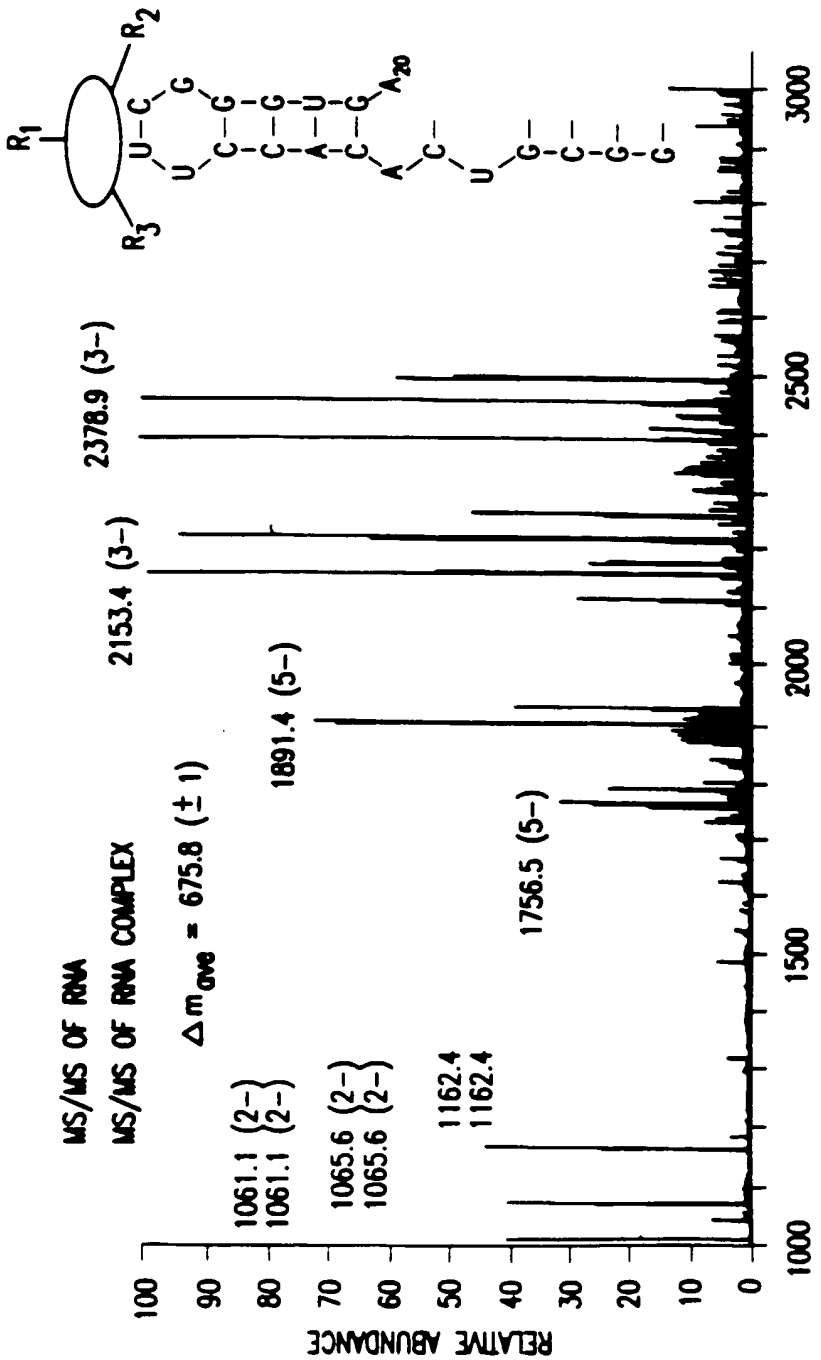

MS/MS OF RNA
MS/MS OF RNA COMPLEX
Δm one = 675.8 (± 1)
2153.4 (3-)
2378.9 (3-)
1891.4 (5-)
1756.5 (5-)
1061.1 (2-)
1061.1 (2-)
1065.6 (2-)
1065.6 (2-)
1162.4
1162.4
RELATIVE ABUNDANCE
m/z
100
90
80
70
60
50
40
30
20
10
0
1000
1500
2000
2500
3000

Figure 49

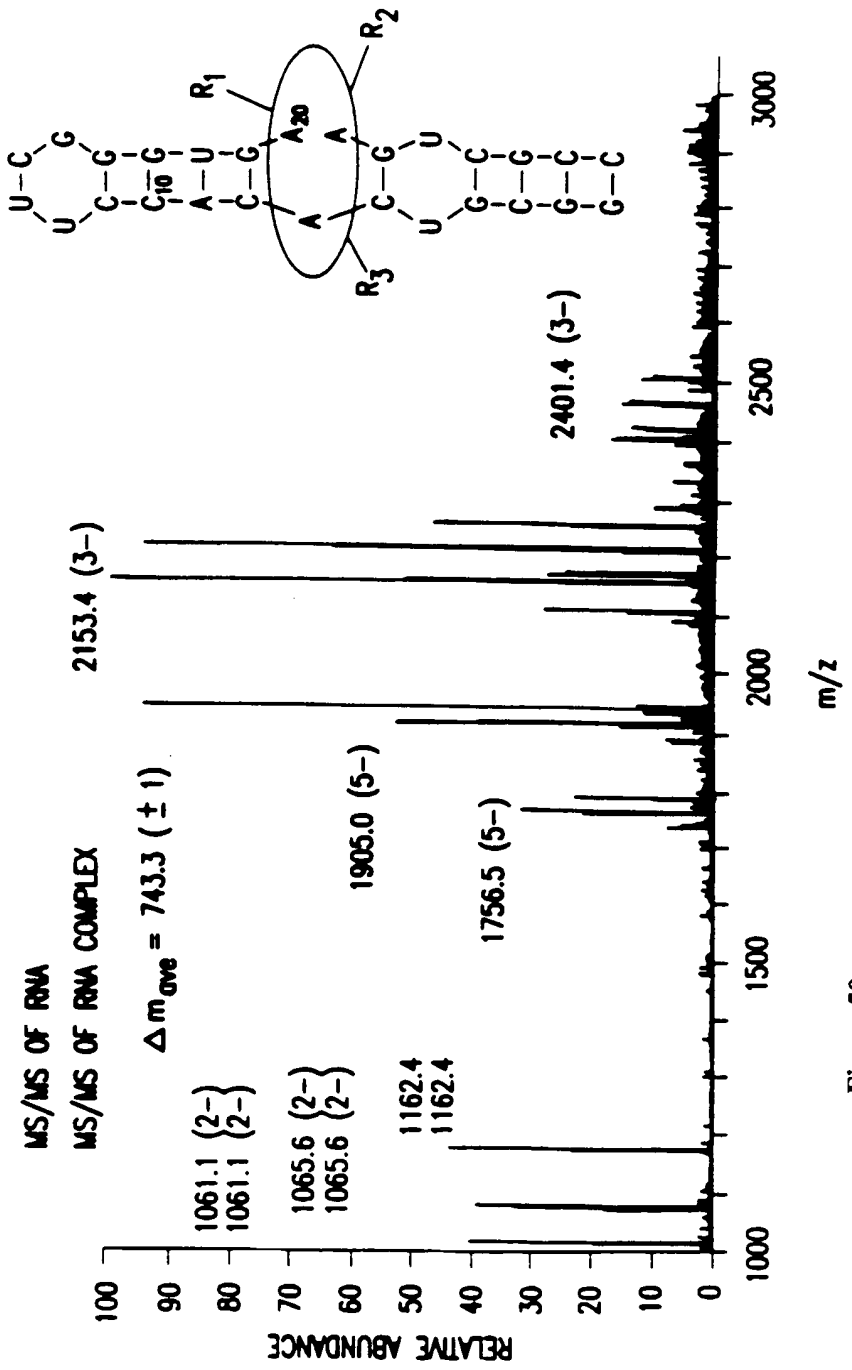

MS/MS OF RNA
MS/MS OF RNA COMPLEX
Δm_{ave} = 743.3 (± 1)
2153.4 (3-)
2401.4 (3-)
1905.0 (5-)
1756.5 (5-)
1061.1 (2-)
1061.1 (2-)
1065.6 (2-)
1065.6 (2-)
1162.4
1162.4
RELATIVE ABUNDANCE
m/z
100
90
80
70
60
50
40
30
20
10
0
1000
1500
2000
2500
3000
R_1
R_2
R_3
A_{20}
C_{10}

Figure 50

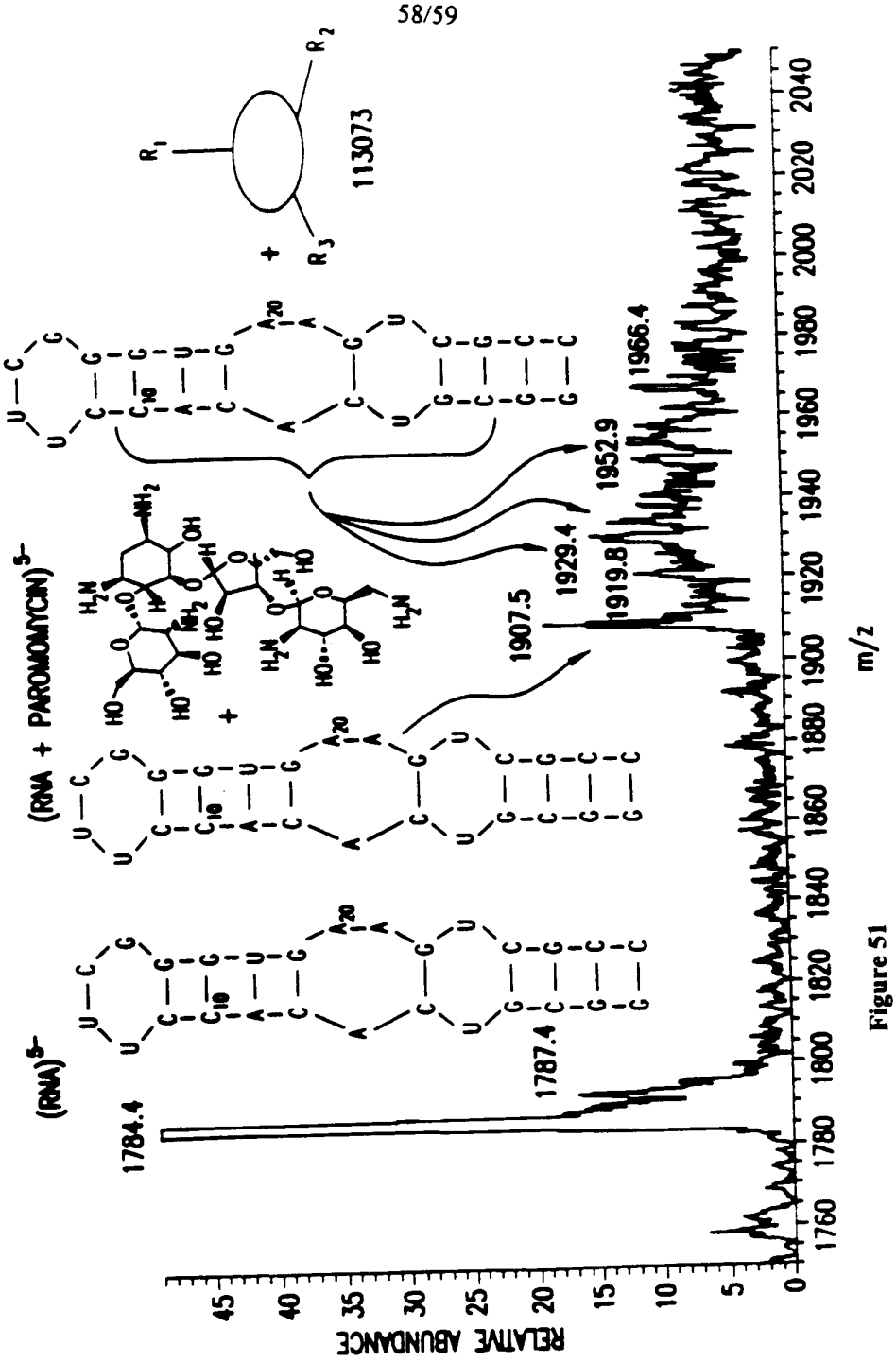

113073
(RNA + PAROMOMYCIN)5-
(RNA)5-
1784.4
1787.4
1907.5
1919.8
1929.4
1952.9
1966.4
RELATIVE ABUNDANCE
m/z

Figure 51

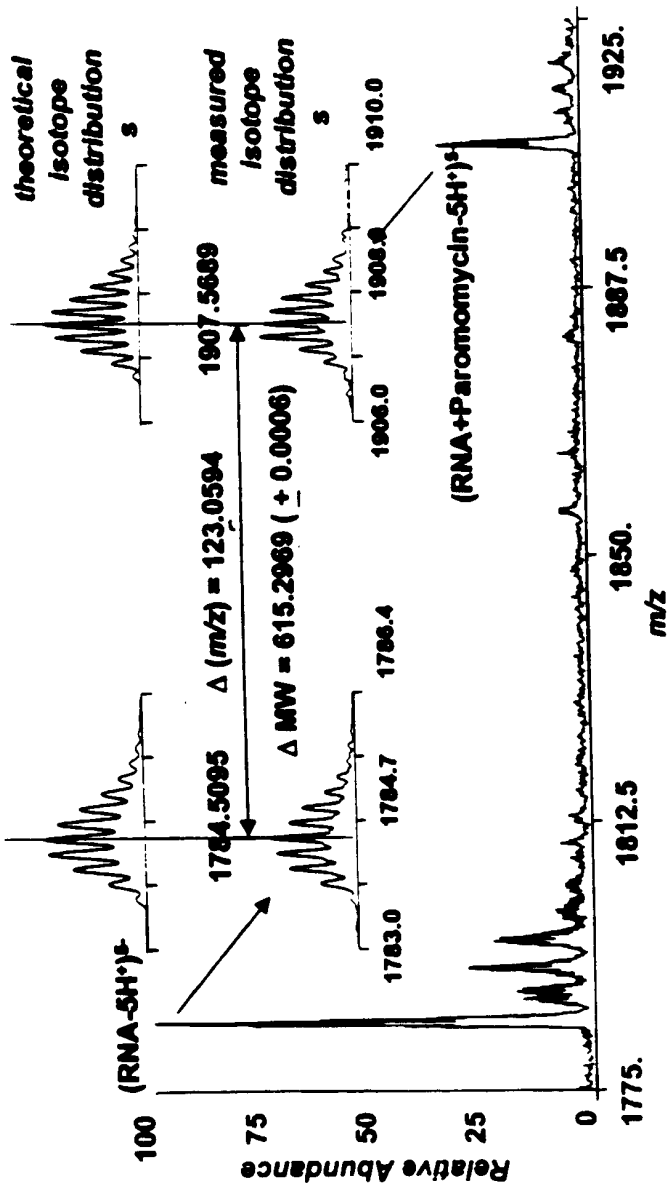

theoretical isotope distribution
measured isotope distribution
(RNA-5H+)5-
(RNA+Paromomycin-5H+)5-
Δ (m/z) = 123.0594
Δ MW = 615.2969 (± 0.0006)
1907.5689
1910.0
1908.8
1906.0
1904.5095
1906.4
1904.7
1903.0
100
75
50
25
0
Relative Abundance
1775.
1812.5
1850.
1887.5
1925.
m/z

Figure 52

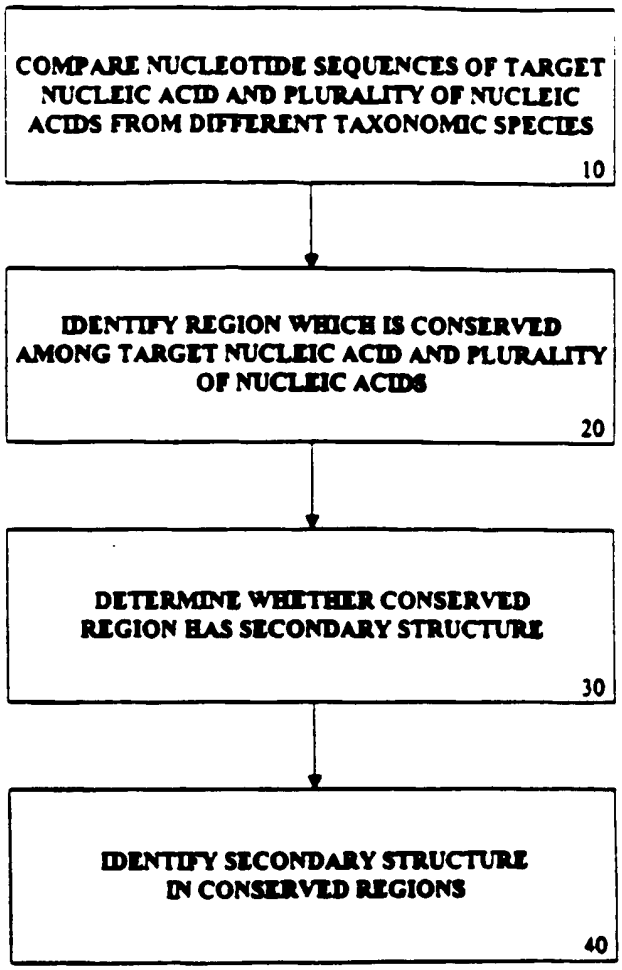

COMPARE NUCLEOTIDE SEQUENCES OF TARGET NUCLEIC ACID AND PLURALITY OF NUCLEIC ACIDS FROM DIFFERENT TAXONOMIC SPECIES
10
IDENTIFY REGION WHICH IS CONSERVED AMONG TARGET NUCLEIC ACID AND PLURALITY OF NUCLEIC ACIDS
20
DETERMINE WHETHER CONSERVED REGION HAS SECONDARY STRUCTURE
30
IDENTIFY SECONDARY STRUCTURE IN CONSERVED REGIONS
40

FIGURE 1

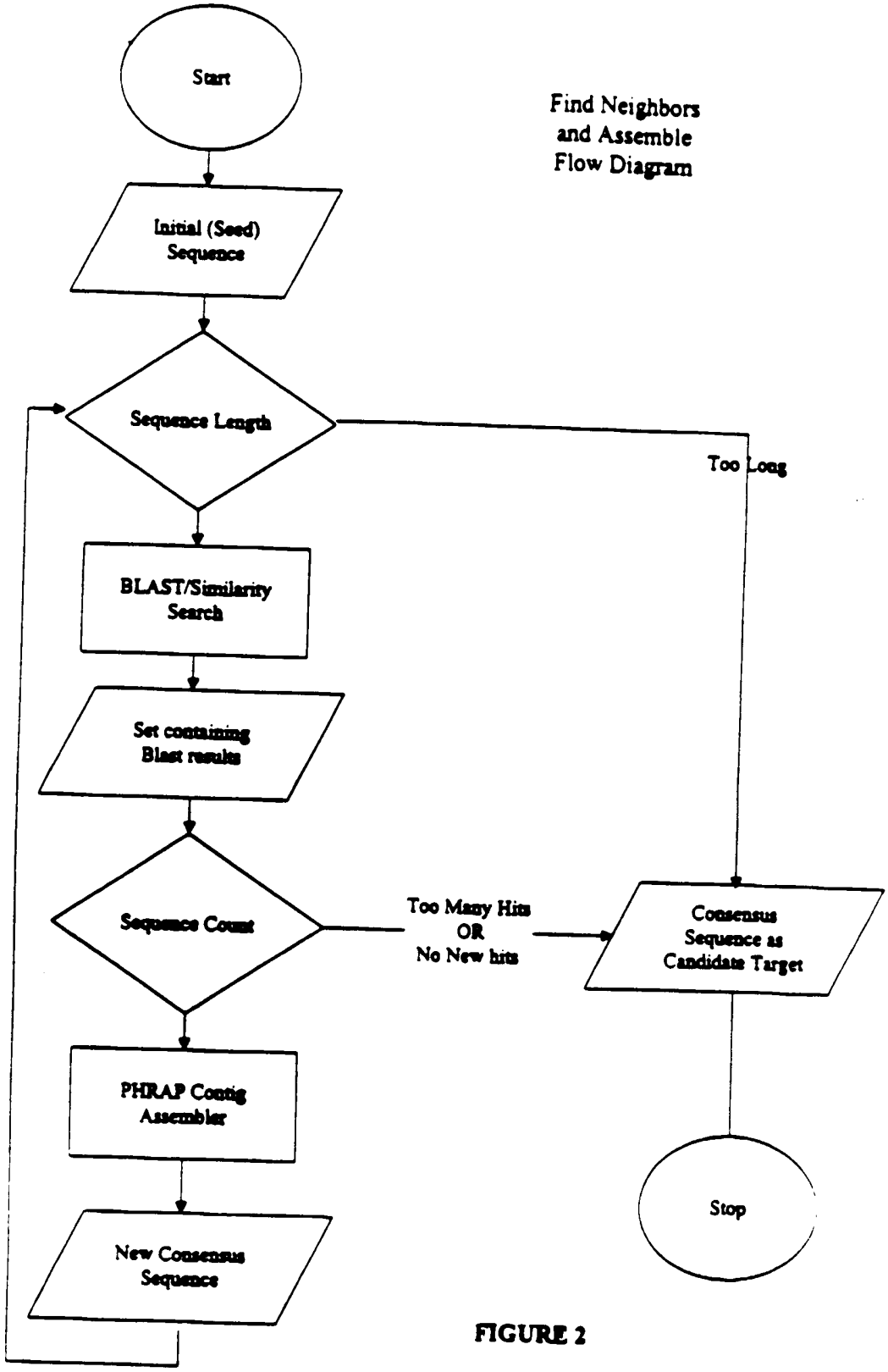

Start
Find Neighbors and Assemble Flow Diagram
Initial (Seed) Sequence
Sequence Length
Too Long
BLAST/Similarity Search
Set containing Blast results
Sequence Count
Too Many Hits OR No New hits
Consensus Sequence as Candidate Target
PHRAP Contig Assembler
Stop
New Consensus Sequence

FIGURE 2

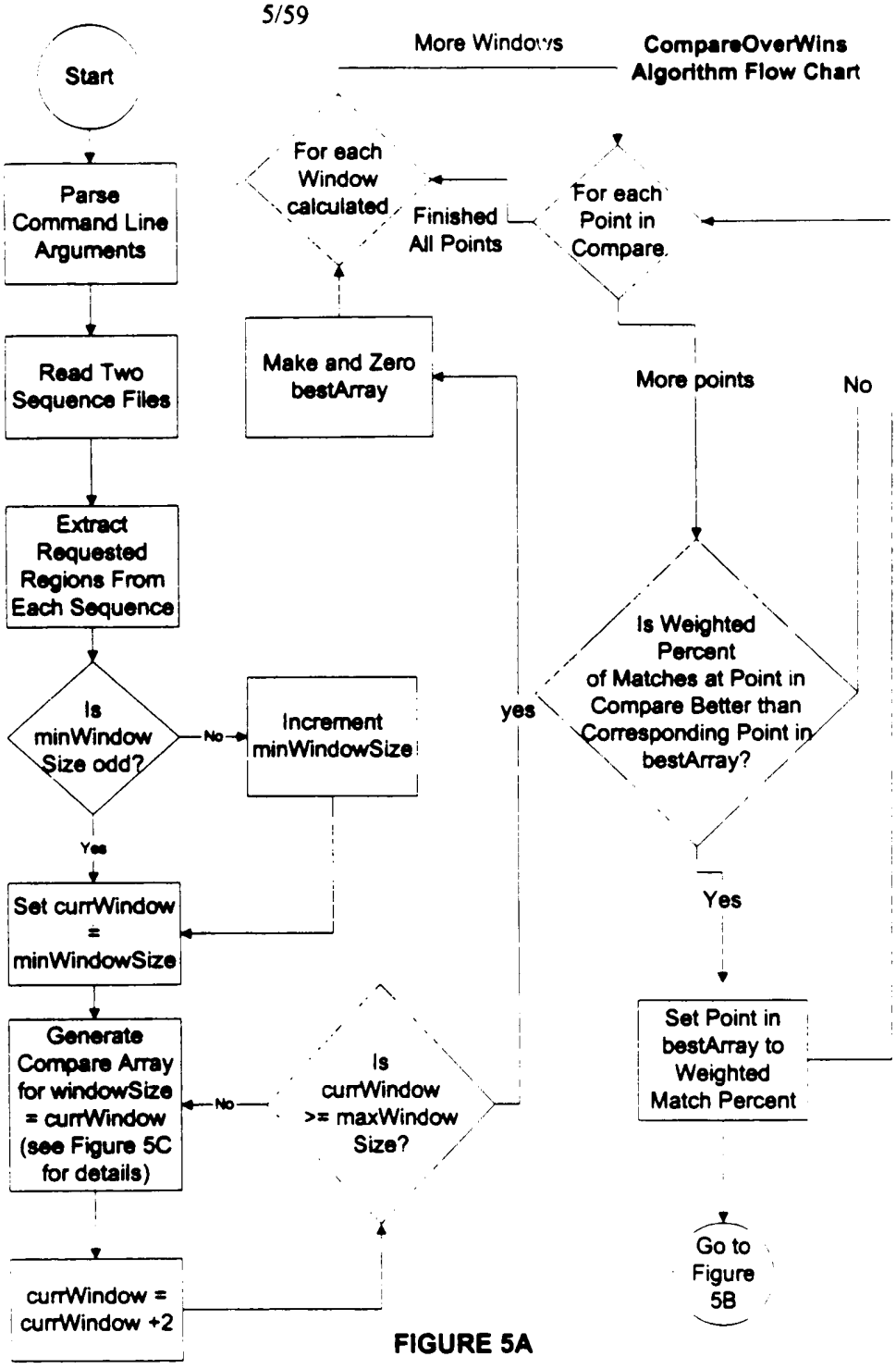

CompareOverWins
Algorithm Flow Chart
Start
Parse Command Line Arguments
Read Two Sequence Files
Extract Requested Regions From Each Sequence
Is minWindow Size odd?
No
Increment minWindowSize
Yes
Set currWindow = minWindowSize
Generate Compare Array for windowSize = currWindow (see Figure 5C for details)
currWindow = currWindow +2
Is currWindow >= maxWindow Size?
No
yes
Make and Zero bestArray
For each Window calculated
Finished All Points
More Windows
For each Point in Compare
More points
No
Is Weighted Percent of Matches at Point in Compare Better than Corresponding Point in bestArray?
Yes
Set Point in bestArray to Weighted Match Percent
Go to Figure 5B

FIGURE 5A

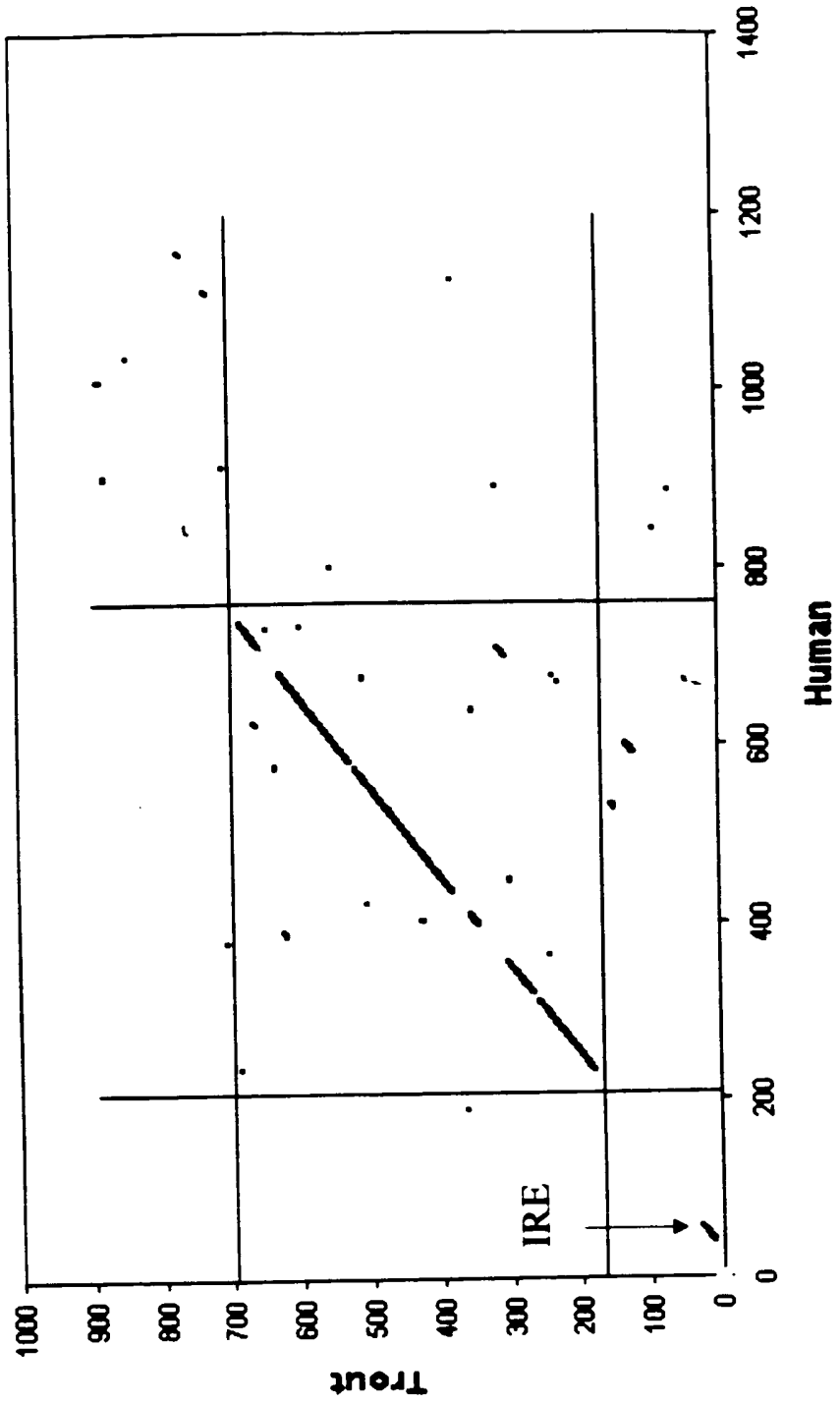

Human
Trout
IRE
0
200
400
600
800
1000
1200
1400
100
200
300
400
500
600
700
800
900
1000

Figure 11

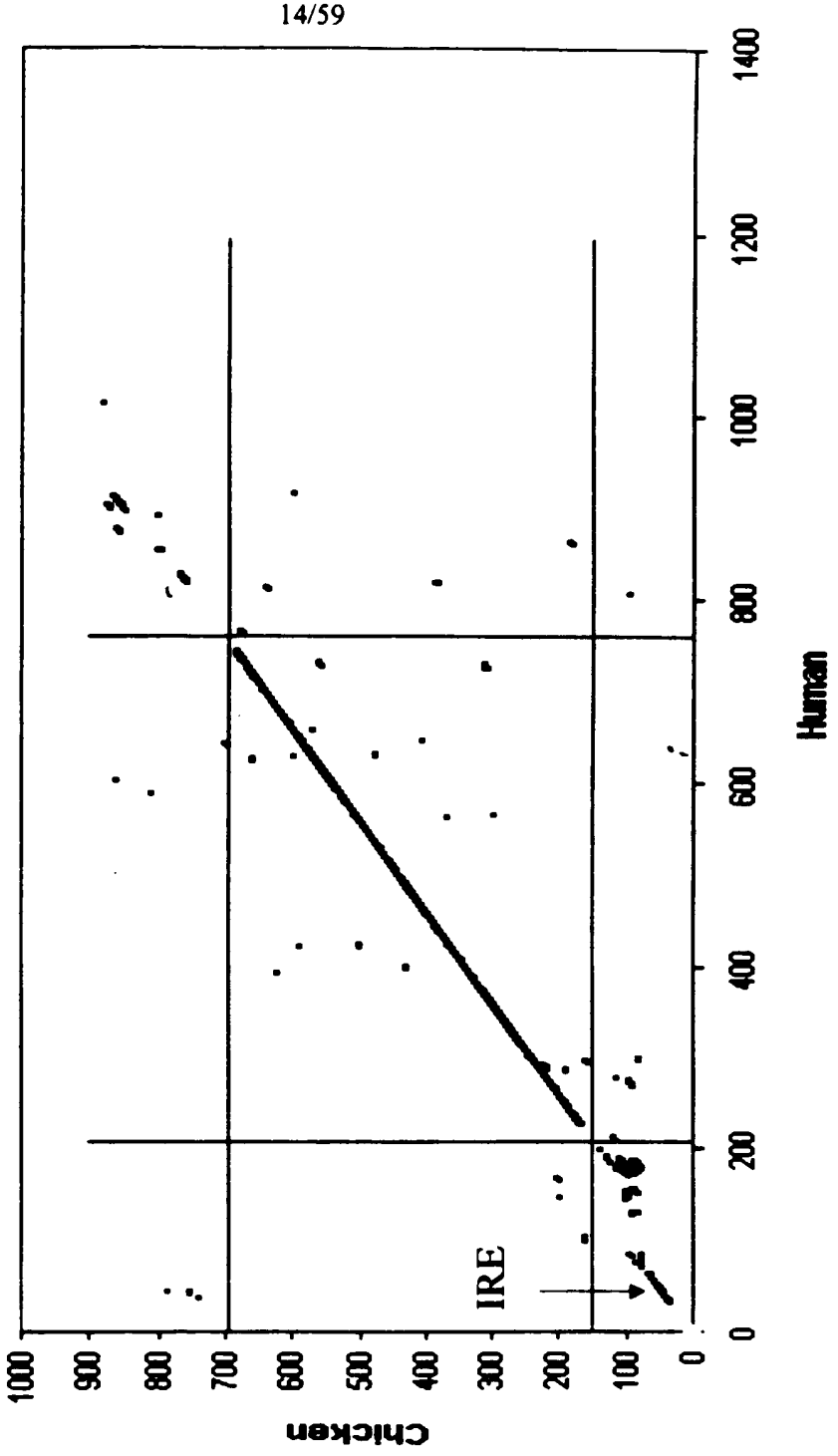
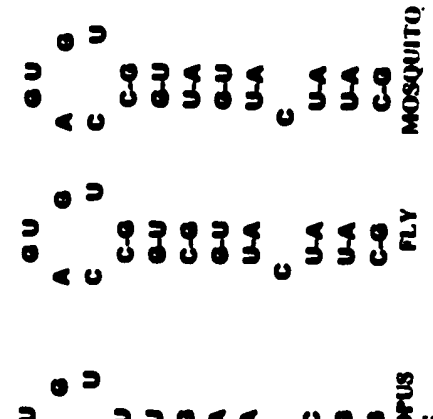

Chicken
1000
900
800
700
600
500
400
300
200
100
0
Human
0
200
400
600
800
1000
1200
1400
IRE

Figure 12

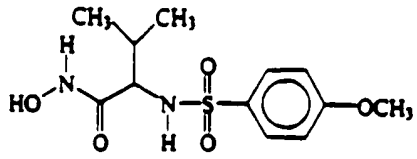


	HUMAN PIG	HAMSTER MOUSE RAT	CHICKEN	TROUT SALMON	XENOPUS FROG	FLY	MOSQUITO
HUMAN PIG	No	No	Yes	Yes	Yes	No	No
HAMSTER MOUSE RAT		No	Yes	Yes	Yes	No	No
CHICKEN			No	Yes	Yes	No	No
TROUT SALMON				No	Yes	Yes	Yes
XENOPUS FROG					No	Yes	Yes
FLY						No	Yes
MOSQUITO							No

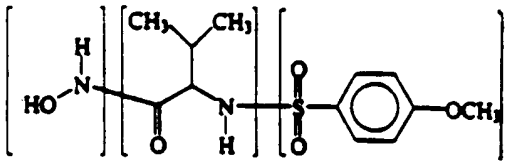
Figure 13

16/59

Compound CI



$C_{12}H_{18}N_2O_5S$



	F_i	F_{ii}	F_{iii}
Molecular formula	H_2NO	C_5H_9NO	$C_7H_7O_3S$

Figure 14

Addition of fragments to yield compounds

Table

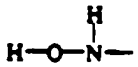
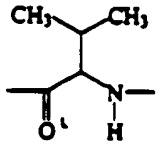
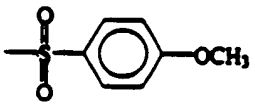
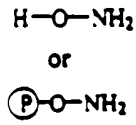
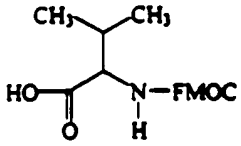
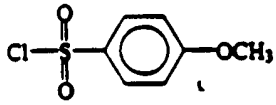
Fragment Identifier	Structure	Name	Molecular formula	Other
F_i		Hydroxylamine	H_2NO	...
F_{ii}		Amino acid	C_5H_9NO	...
F_{iii}		Sulfonyl	$C_7H_8O_3S$	...

Figure 15

Reagents	Identifier	Name	Properties
	R_i	Hydroxylamine	...
	R_{ii}	FMOC blocked amino acid	...
	R_{iii}	Sulfonylchloride	...

Ⓟ = Solid support

Figure 16

Transformation

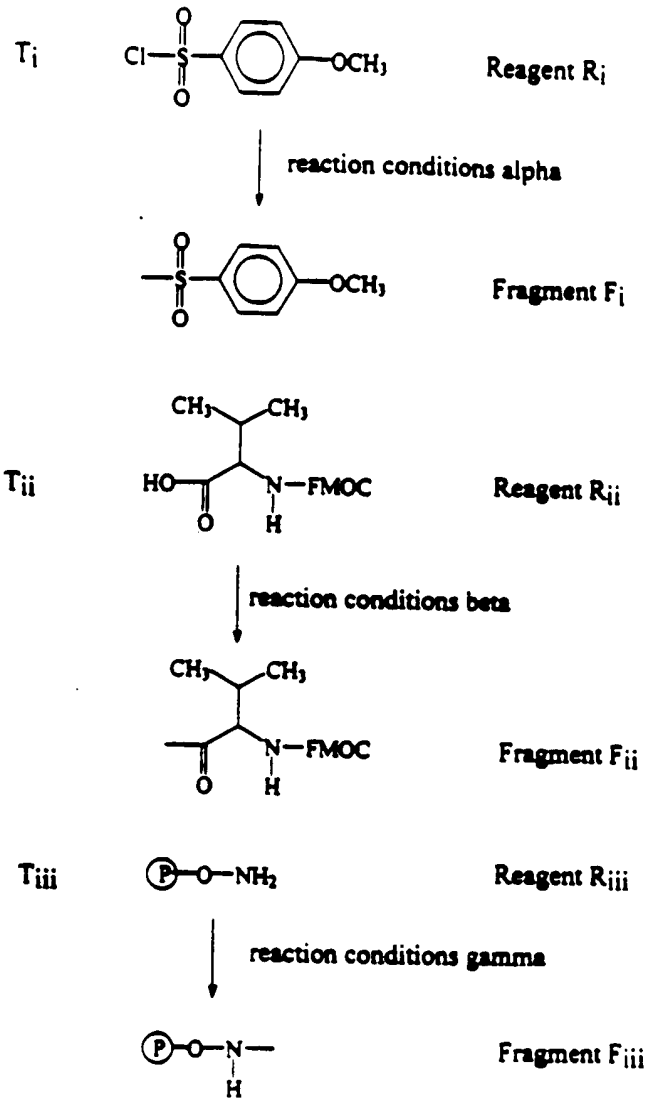


Figure 17

Common Fragment / Different Reagents and Transformations

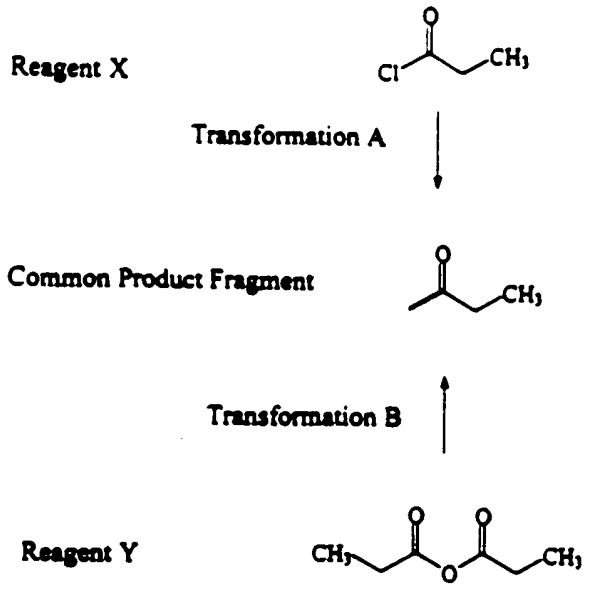


Figure 18

Common Fragment / Different Reagents and Transformations

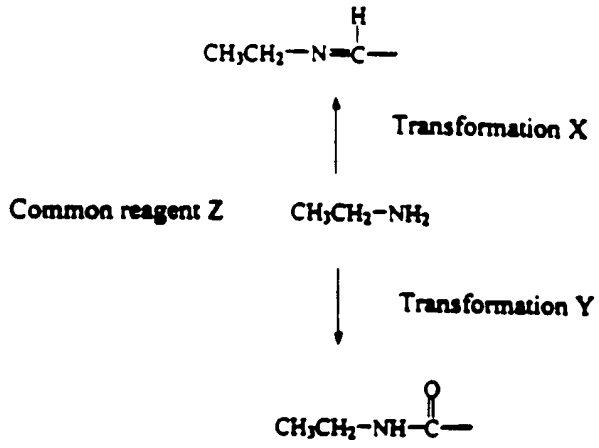


Figure 19A

Common Reagent

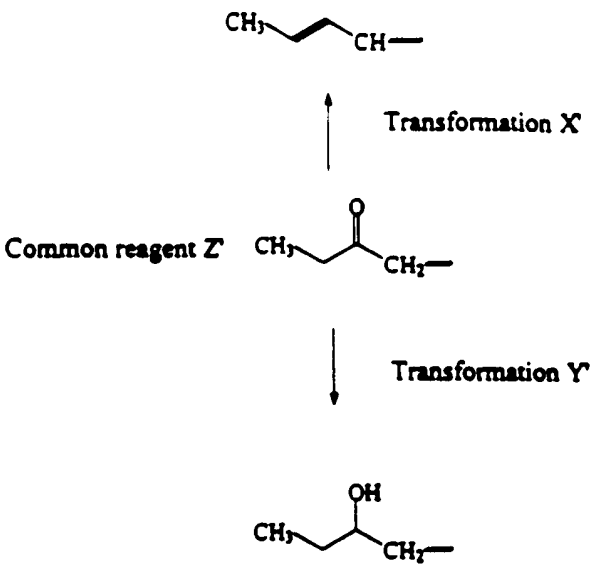
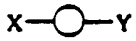
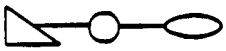


Figure 19B

Symbolic addition of fragments to yield compound

Symbolic Structure	Symbolic Identifier	Molecular formula
Fragment		
	$F_{i'}$	$C_uH_vN_w$ ...
	$F_{ii'}$	$C_uH_vN_w$ ...
	$F_{iii'}$	$C_uH_vN_w$ ...
Compound		
	CI'	$C_uH_vN_w$ ...

Molecular formula $F_{i'}$
+
Molecular formula $F_{ii'}$
+
Molecular formula $F_{iii'}$
= Molecular formula CI'

Figure 20

Symbolic Reagent Table

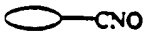
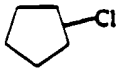
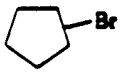
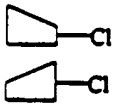
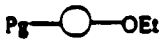
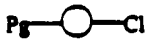
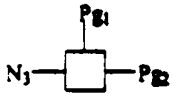
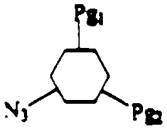
Identifier	Name	Structure	Molecular formula
R1	xxx		xxx
R2	...		...
R3	...		...
R4	...		...
R5	...		...
R6	...		...
R7	...		...
R8	...		...
R9	...		...
R10	...		...

Figure 21

Symbolic Fragment Table

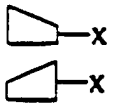
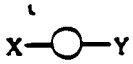
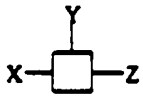
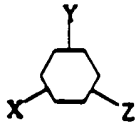
Identifier	Symbolic Structure	Molecular formula	Molecular Weight
F1		xxx	xxx
F2		...	...
F3		...	...
F4		...	...
F5		...	...
F6		...	...
F7		...	...
F8		...	...

Figure 22

Symbolic Transformation Table

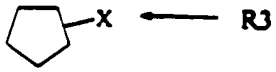
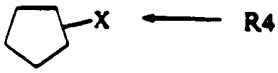
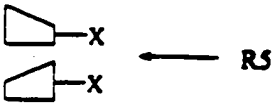
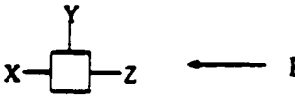
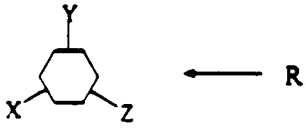
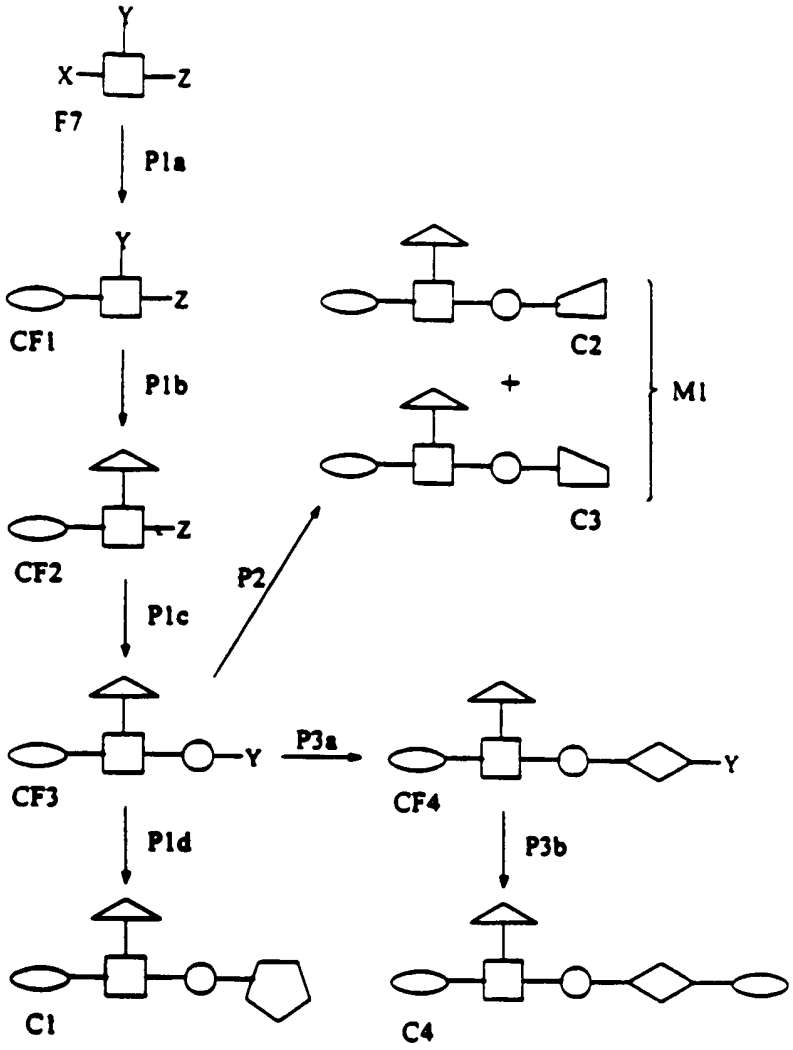
Identifier		Symbolic Reactions		Reagent
T1	F1		R1	conditions α
T2	F2		R2	conditions β
T3	F3		R3	conditions α
T4	F3		R4	conditions α
T5	F4		R5	conditions α
T6	F5		R6	conditions ε
T7	F5		R7	conditions α
T8	F6		R8	conditions α
T9	F7		R9	conditions γ
T10	F8		R10	conditions γ

Figure 23

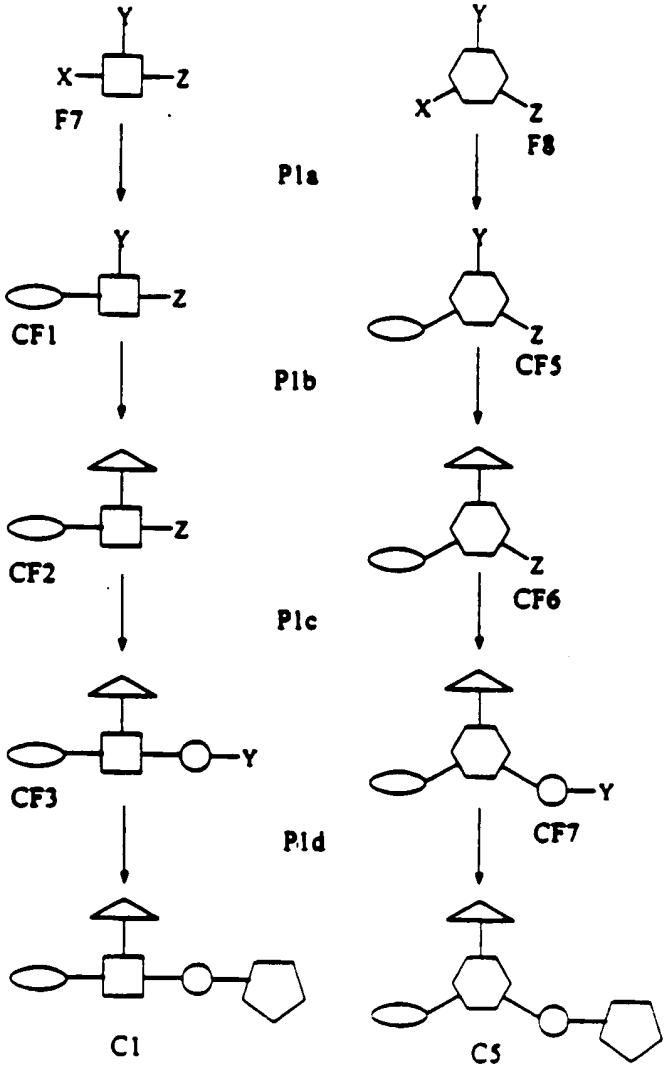
Single Compounds and Mixtures



P = synthetic path CF = complex fragment
F = fragment M = mixture
C = compound

Figure 24

Mixture 2



M2

Figure 25

Mixture 3

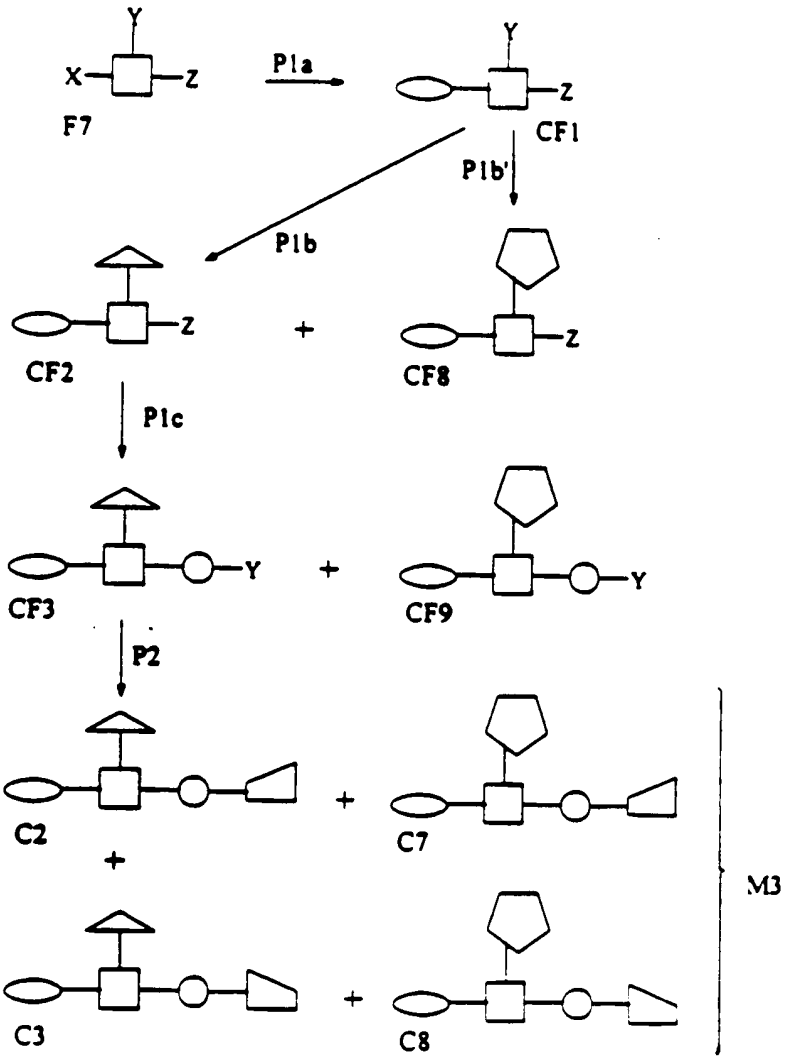
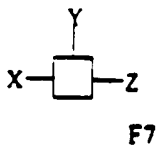


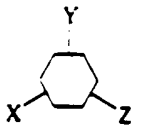
Figure 26

Mixture 4

2 Starting Fragments

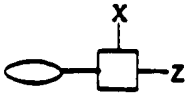


F7

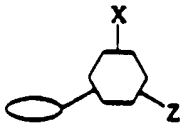


F8

2 Complex Fragments

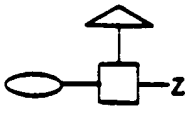


CF1

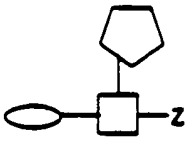


CF5

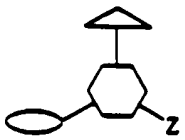
4 Complex Fragments



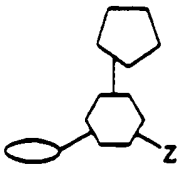
CF2



CF8

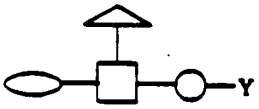


CF 6

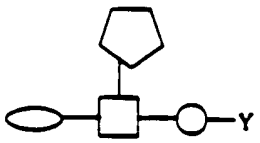


CF11

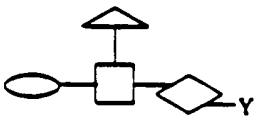
8 Complex Fragments



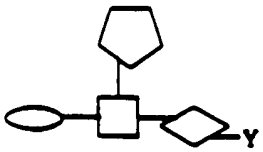
CF3



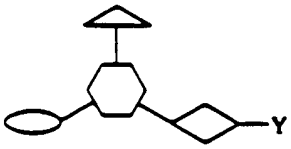
CF9



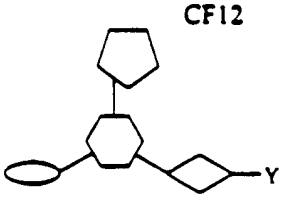
CF12



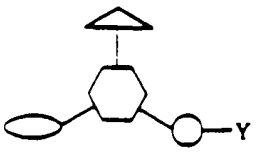
CF13



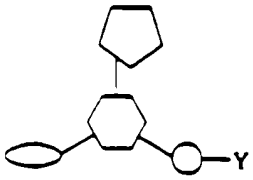
CF15



CF16



CF 7



CF14

Figure 27A

Mixture 4 (continued)

16 compounds

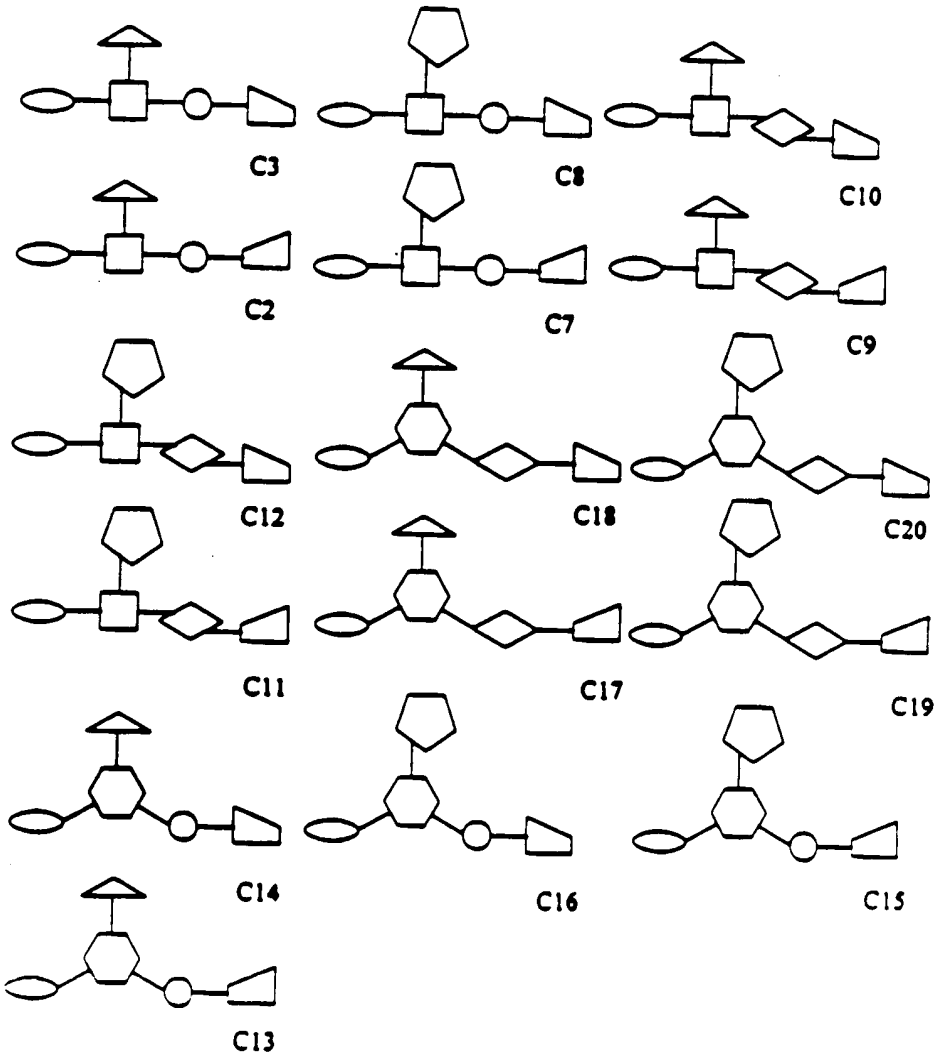
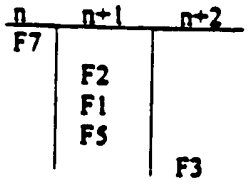


Figure 27B

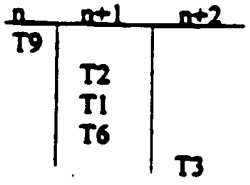
Tracking Table for Compound C1

(a) By Fragments:

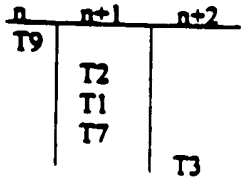


(b) By Transformations:

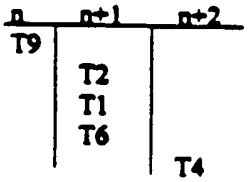
Synthesis Path 1



Synthesis Path 2



Synthesis Path 3



Synthesis Path 4

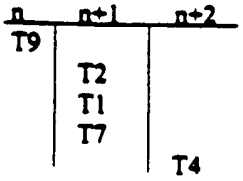


Figure 28

Tracking Table

Tracking M1

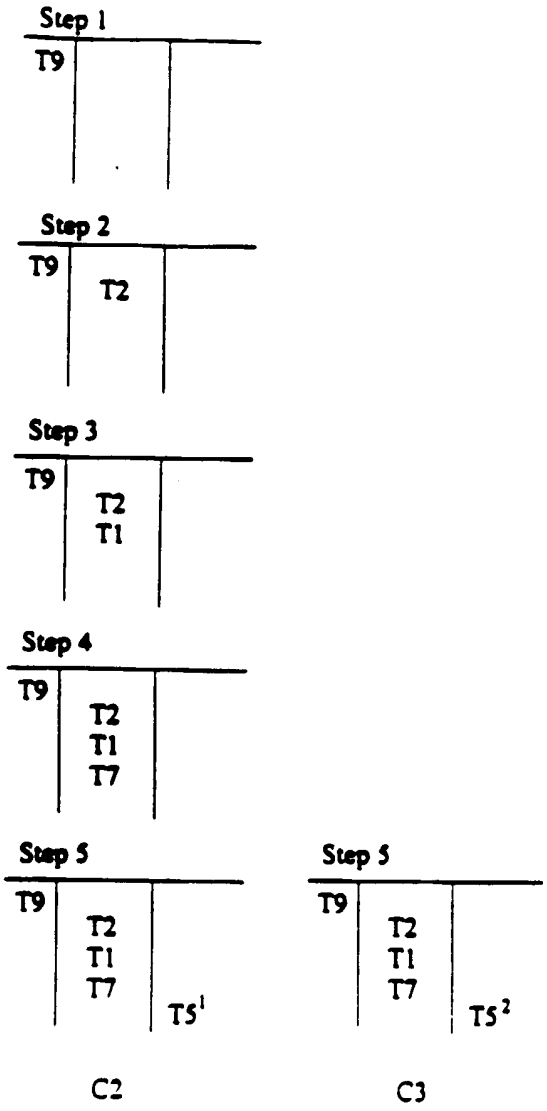


Figure 29

Tracking Table

Tracking M2

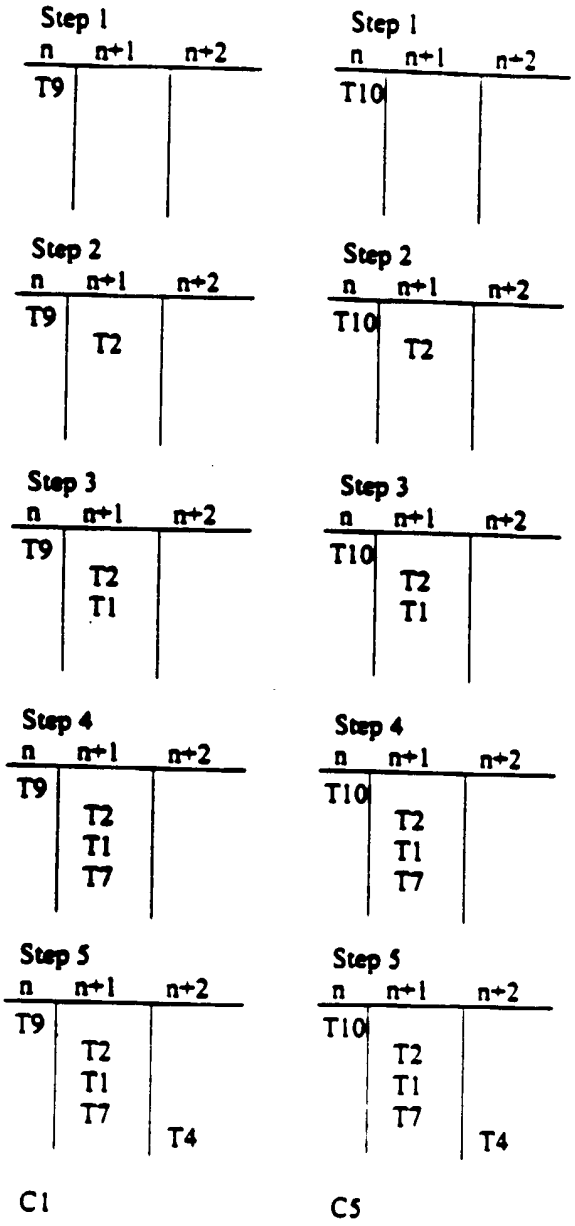


Figure 30

Tracking Table

Tracking M3

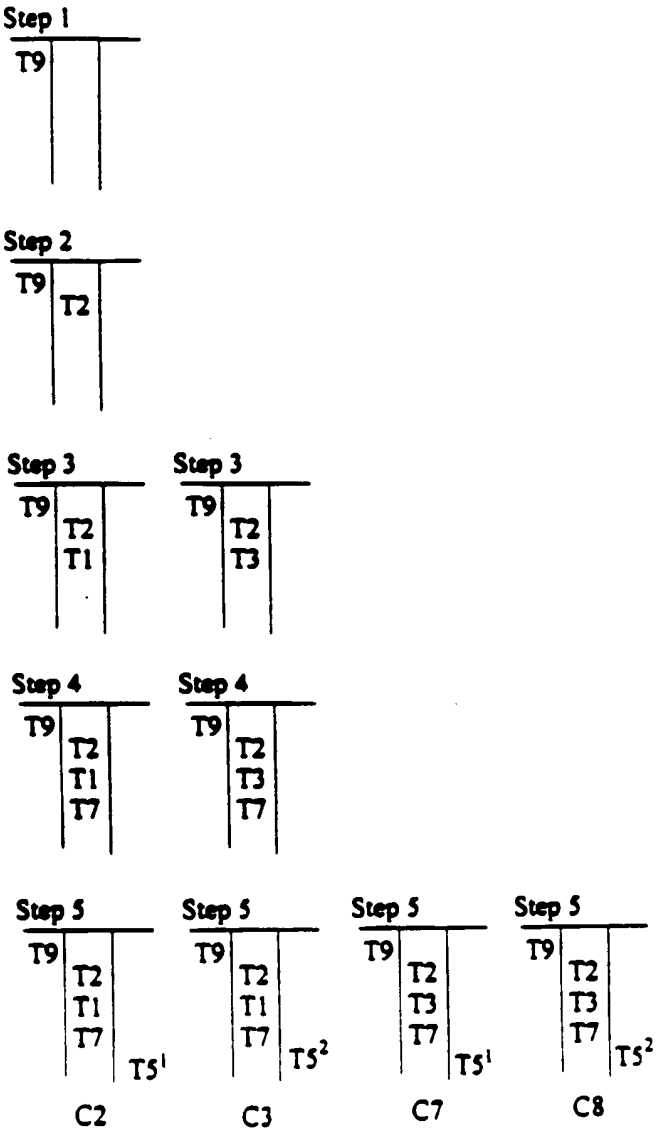


Figure 31

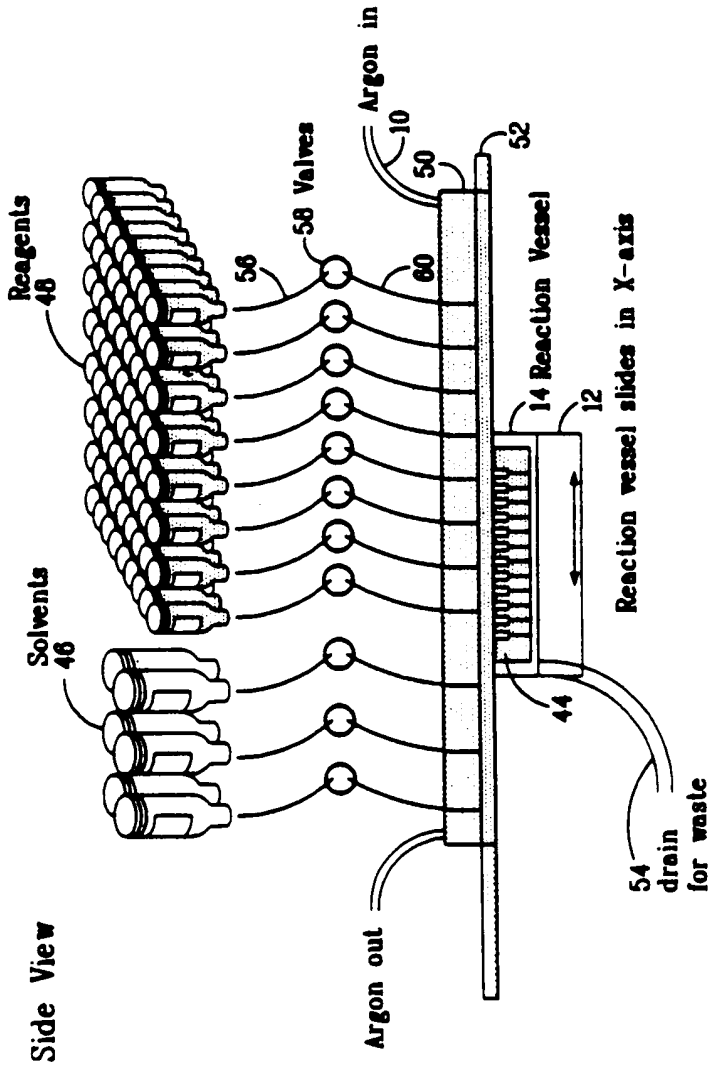

Argon in
10
50
52
58 Valves
56
60
Reagents
48
14 Reaction Vessel
12
Reaction vessel slides in X-axis
Solvents
46
44
54
drain
for waste
Argon out
Side View

Figure 32

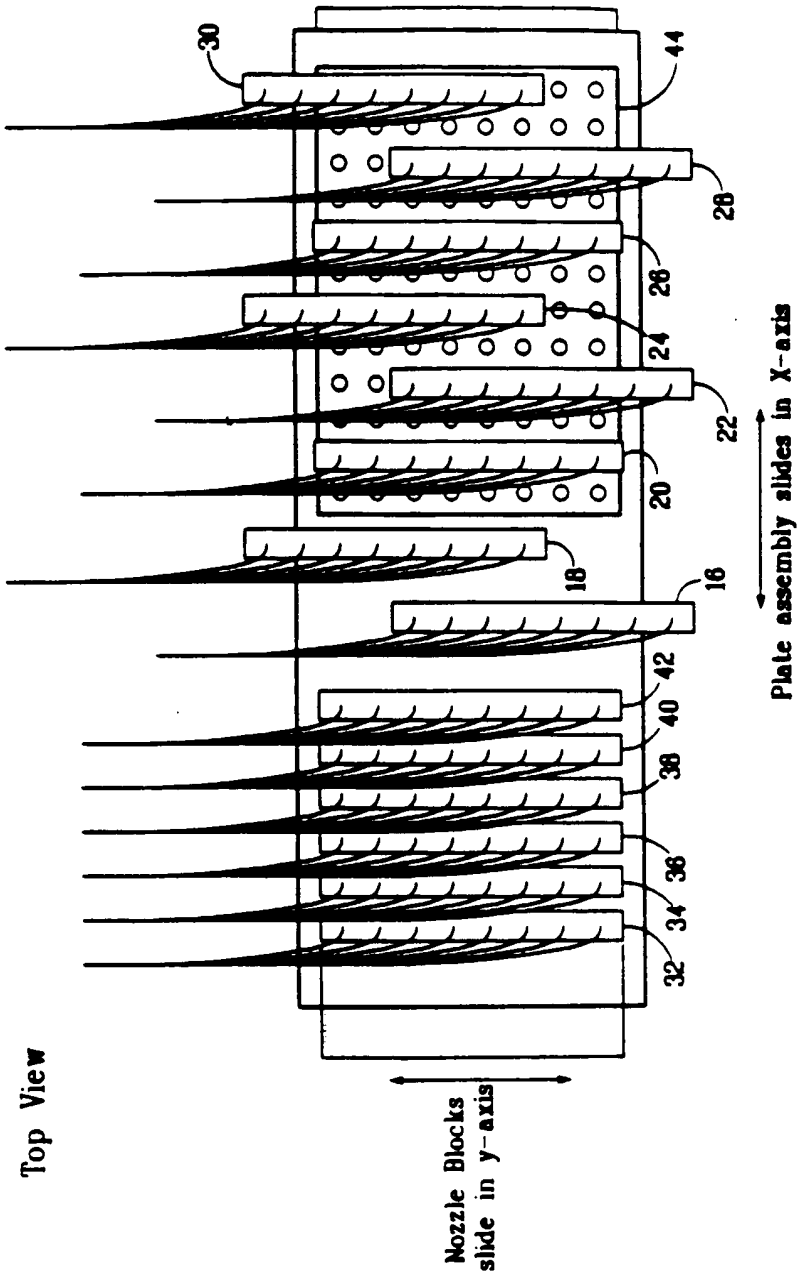

30
44
28
26
24
22
20
18
16
42
40
38
36
34
32
Top View
Nozzle Blocks slide in y-axis
Plate assembly slides in X-axis

Figure 33

Synthesis of hydroxamic acids from alcohol resin

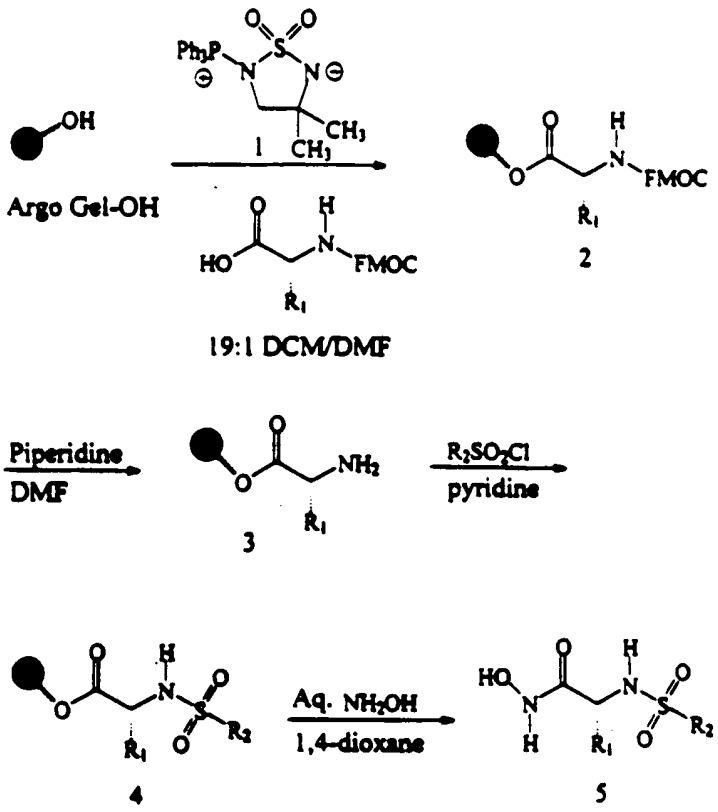


Figure 34

Synthesis of hydroxamic acids from hydroxylamine resin

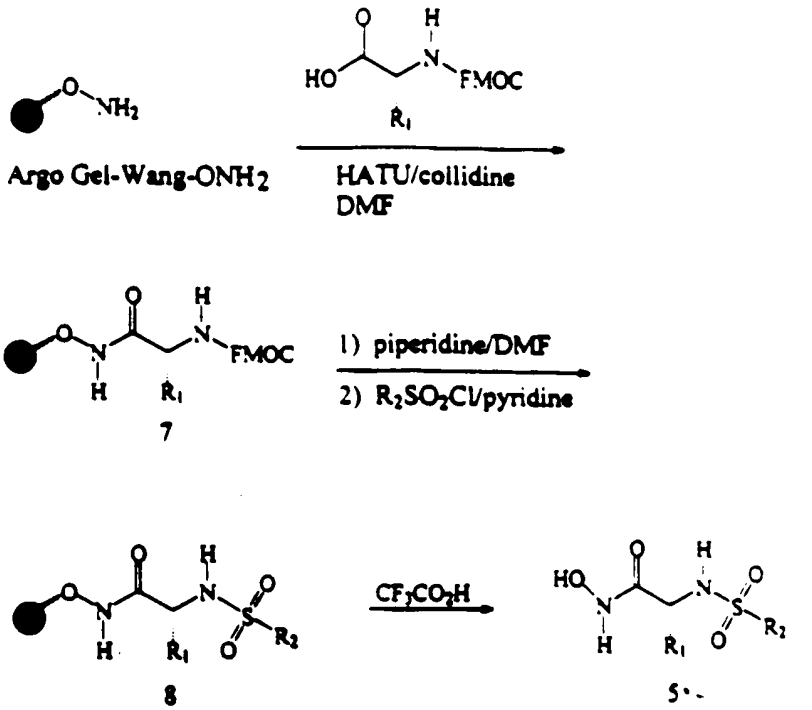


Figure 35

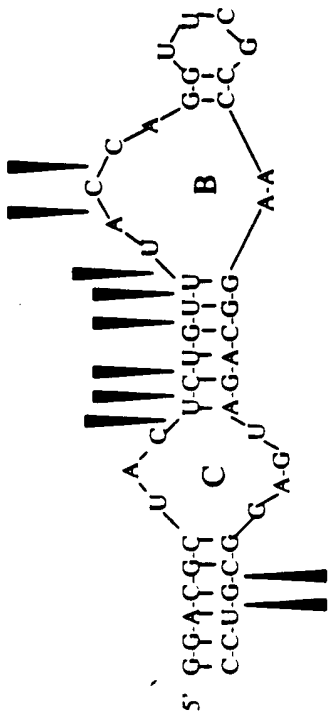


Figure 36

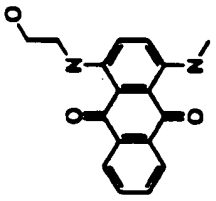
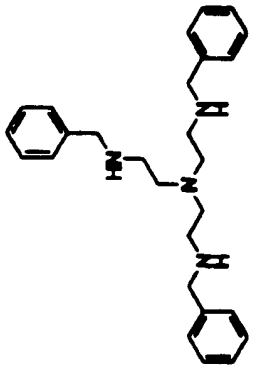
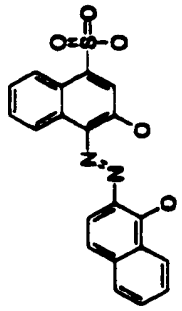
ACD Code	Structure	Calc. ΔG of binding (kcal/mole)	IC_{50} (μM)
00001199		-5.1	< 2
00192509		-8.5	< 2
00003934		-5.1	< 50

Figure 37

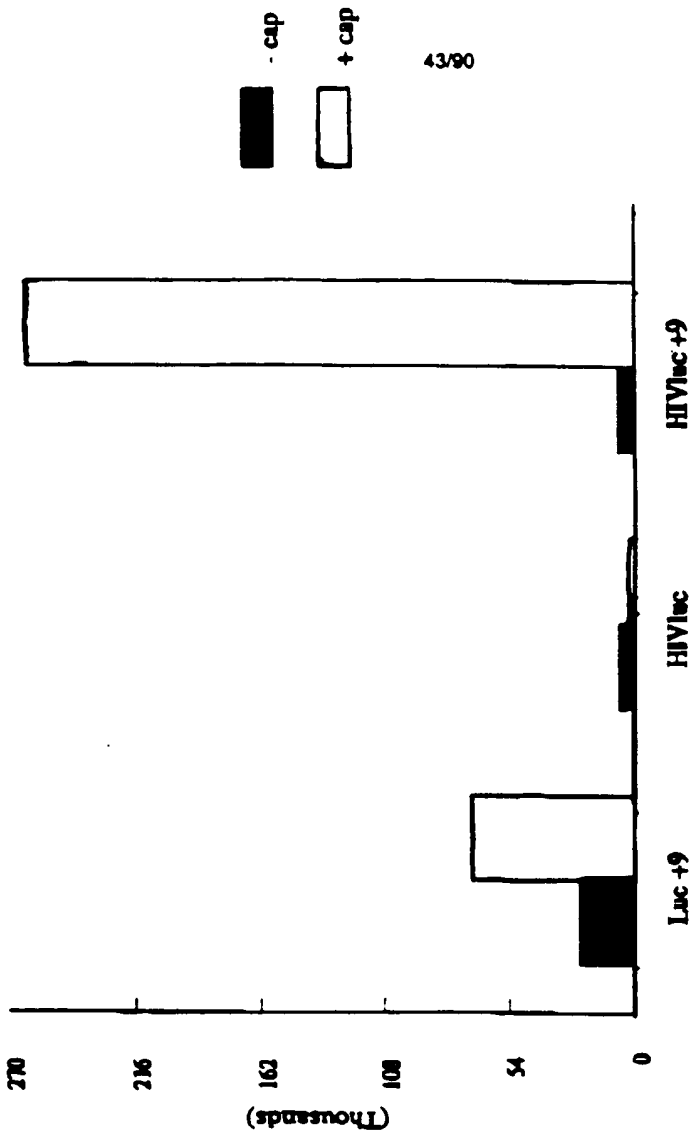

- cap
+ cap
43/90
HIVluc+9
HIVluc
Luc+9
270
216
162
108
54
0
(Thousands)

Figure 38A

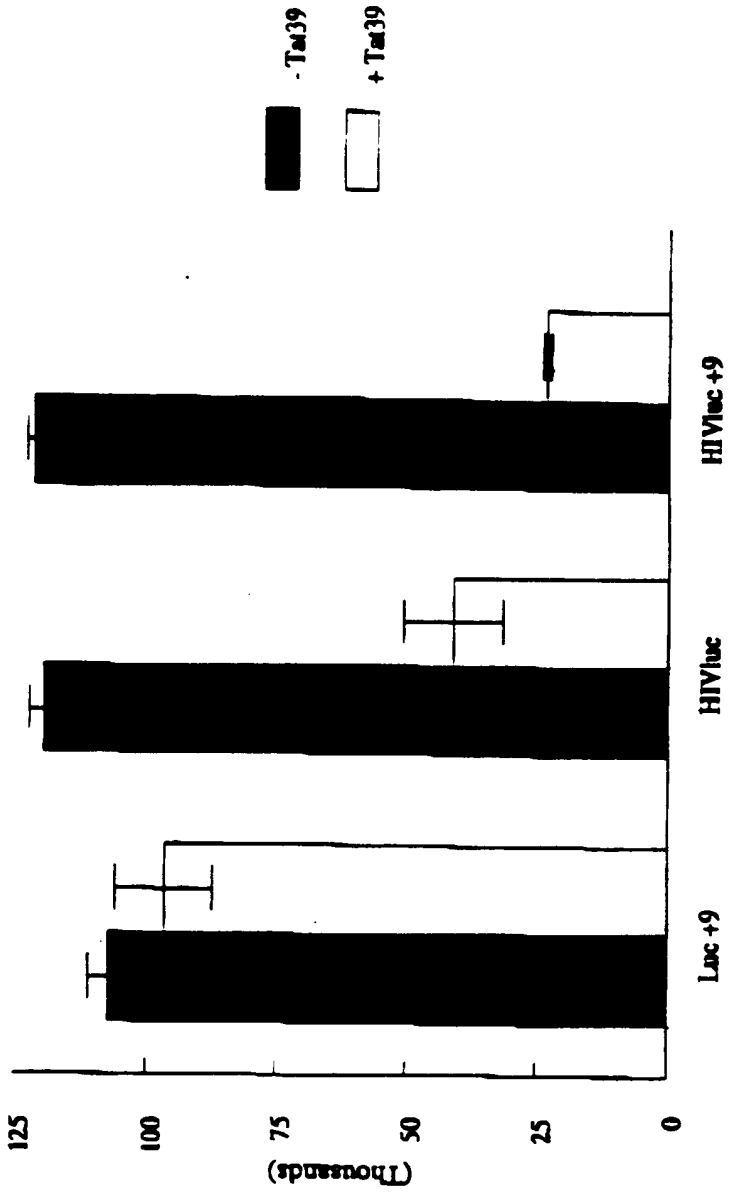


Figure 38B

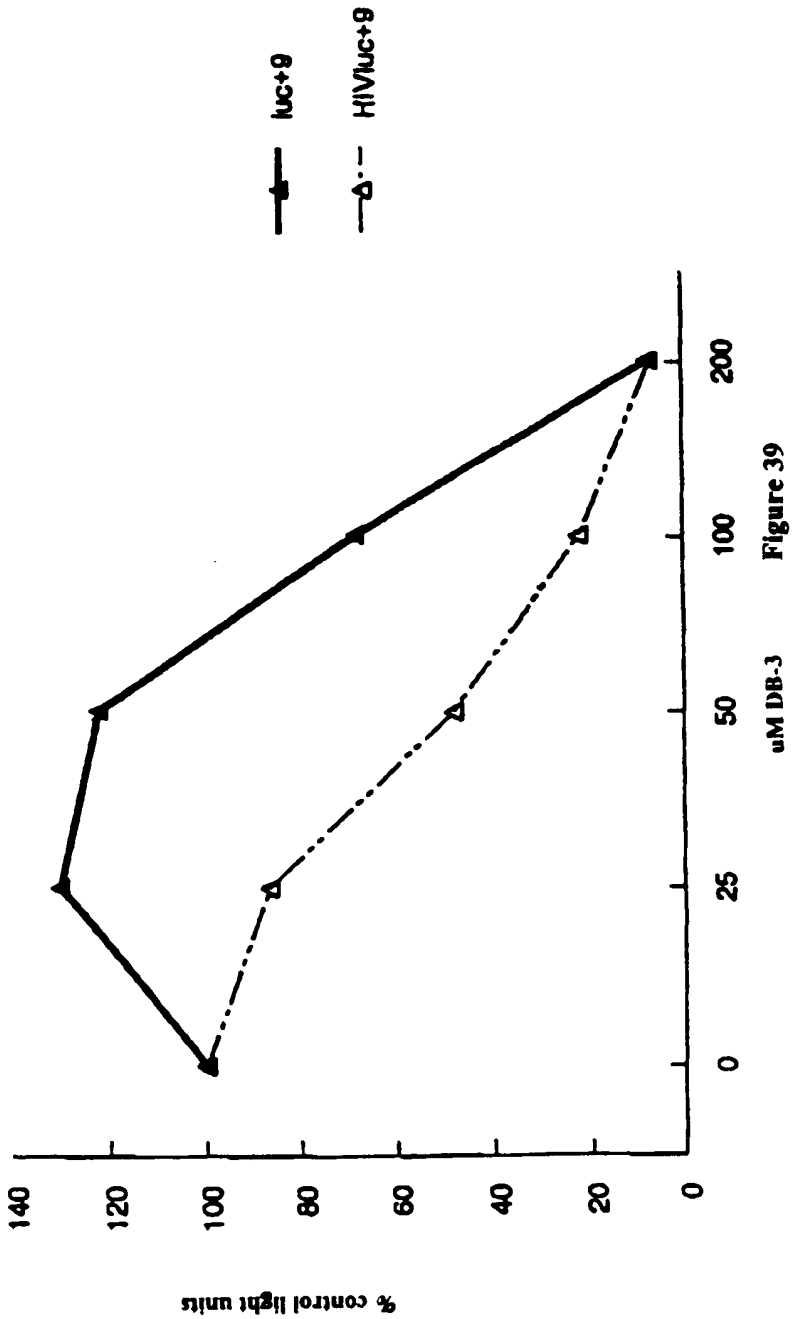

luc+9
HIVluc+9
% control light units
140
120
100
80
60
40
20
0
0
25
50
100
200
uM DB-3

Figure 39

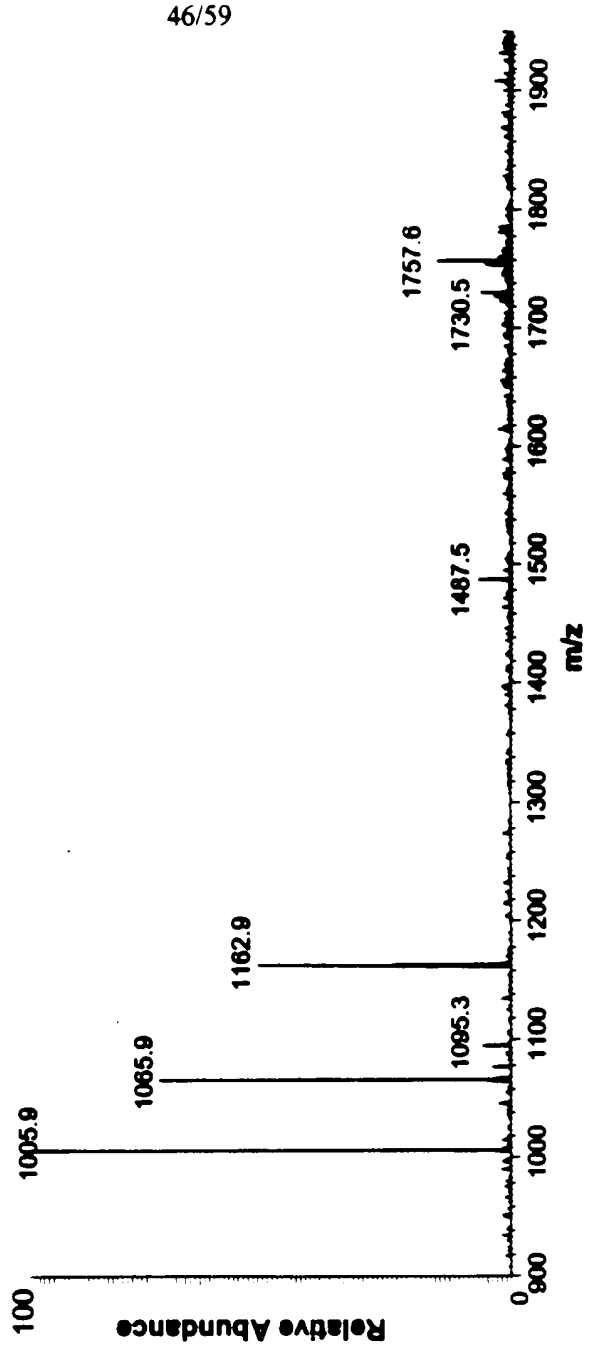

Relative Abundance
100
0
900
1000
1100
1200
1300
1400
1500
1600
1700
1800
1900
m/z
1005.9
1065.9
1085.3
1162.9
1487.5
1730.5
1757.6

Figure 41A

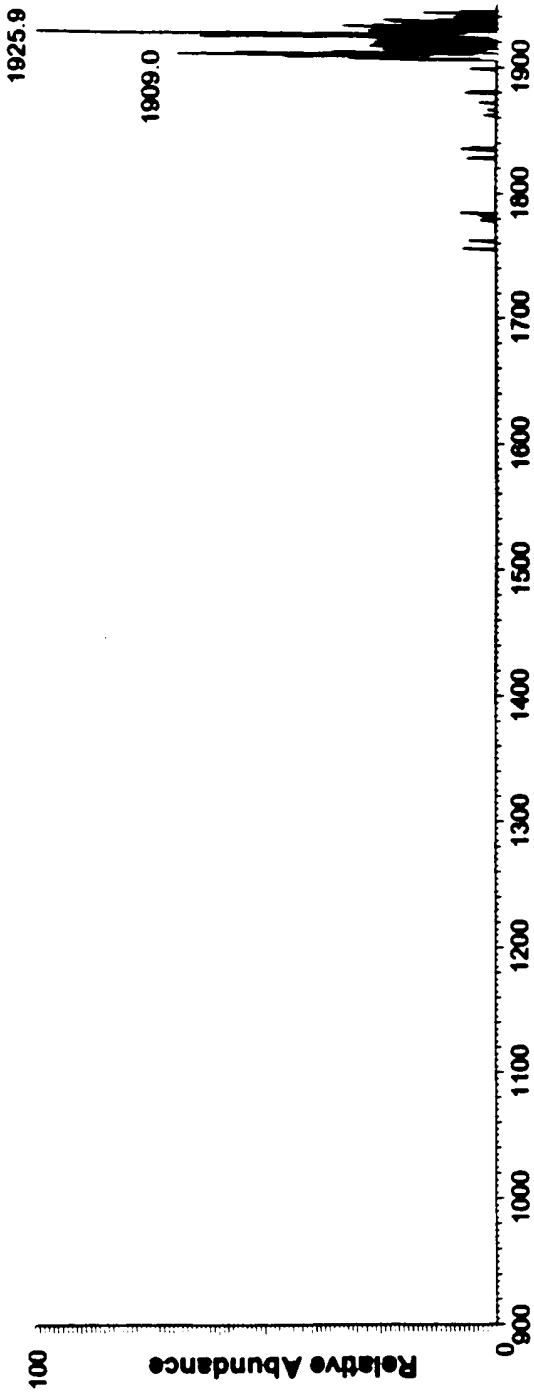

1925.9
1909.0
Relative Abundance
100
0
900
1000
1100
1200
1300
1400
1500
1600
1700
1800
1900
m/z

Figure 41B

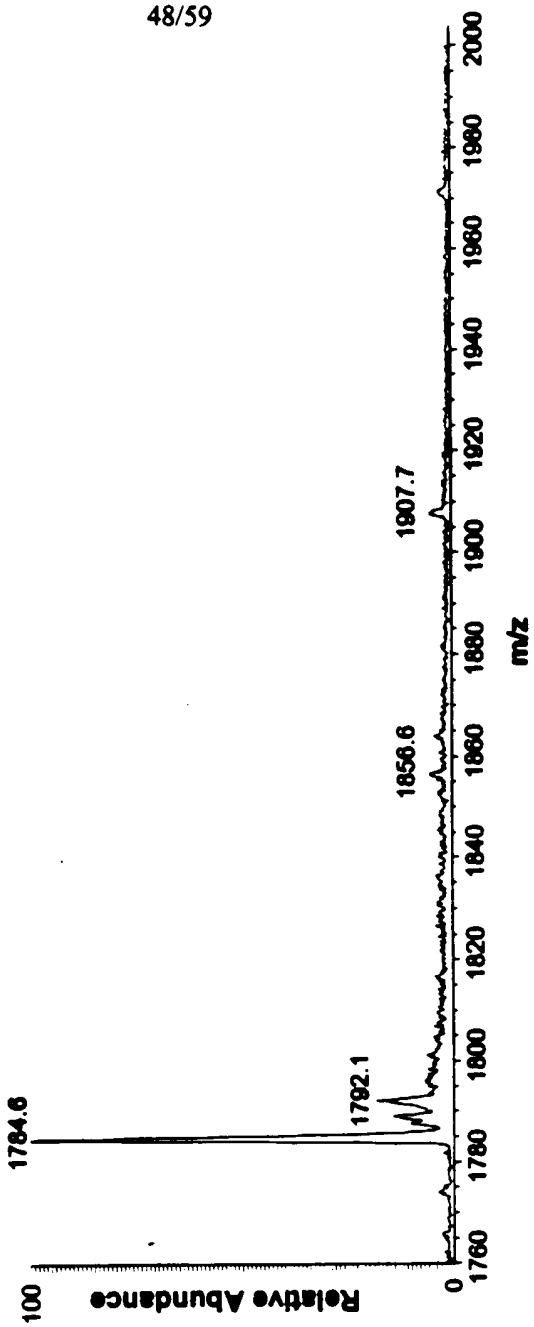

Relative Abundance
100
0
1760
1780
1800
1820
1840
1860
1880
1900
1920
1940
1960
1980
2000
m/z
1784.6
1792.1
1856.6
1907.7

Figure 42A

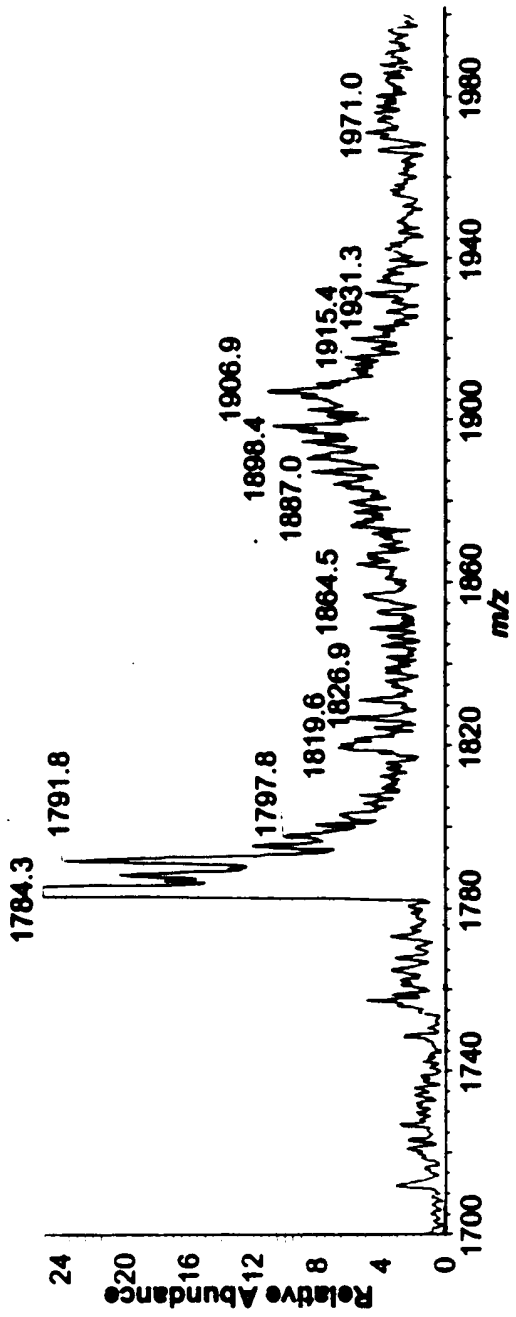

Relative Abundance
m/z
1784.3
1791.8
1797.8
1819.6
1826.9
1864.5
1887.0
1898.4
1906.9
1915.4
1931.3
1971.0
0
4
8
12
16
20
24
1700
1740
1780
1820
1860
1900
1940
1980

Figure 42B

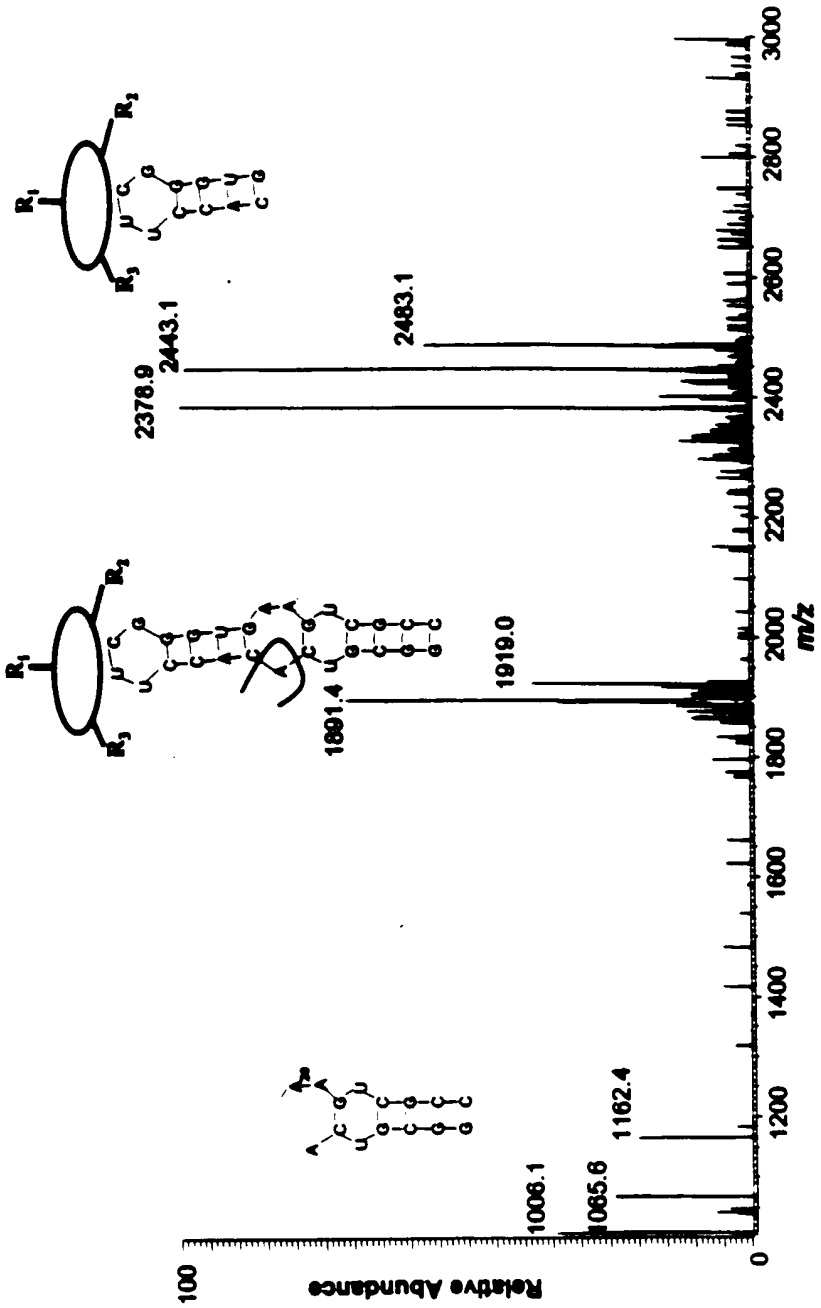

Relative Abundance
100
0
1200
1400
1600
1800
2000
2200
2400
2600
2800
3000
m/z
1006.1
1065.6
1162.4
1891.4
1919.0
2378.9
2443.1
2483.1

Figure 43

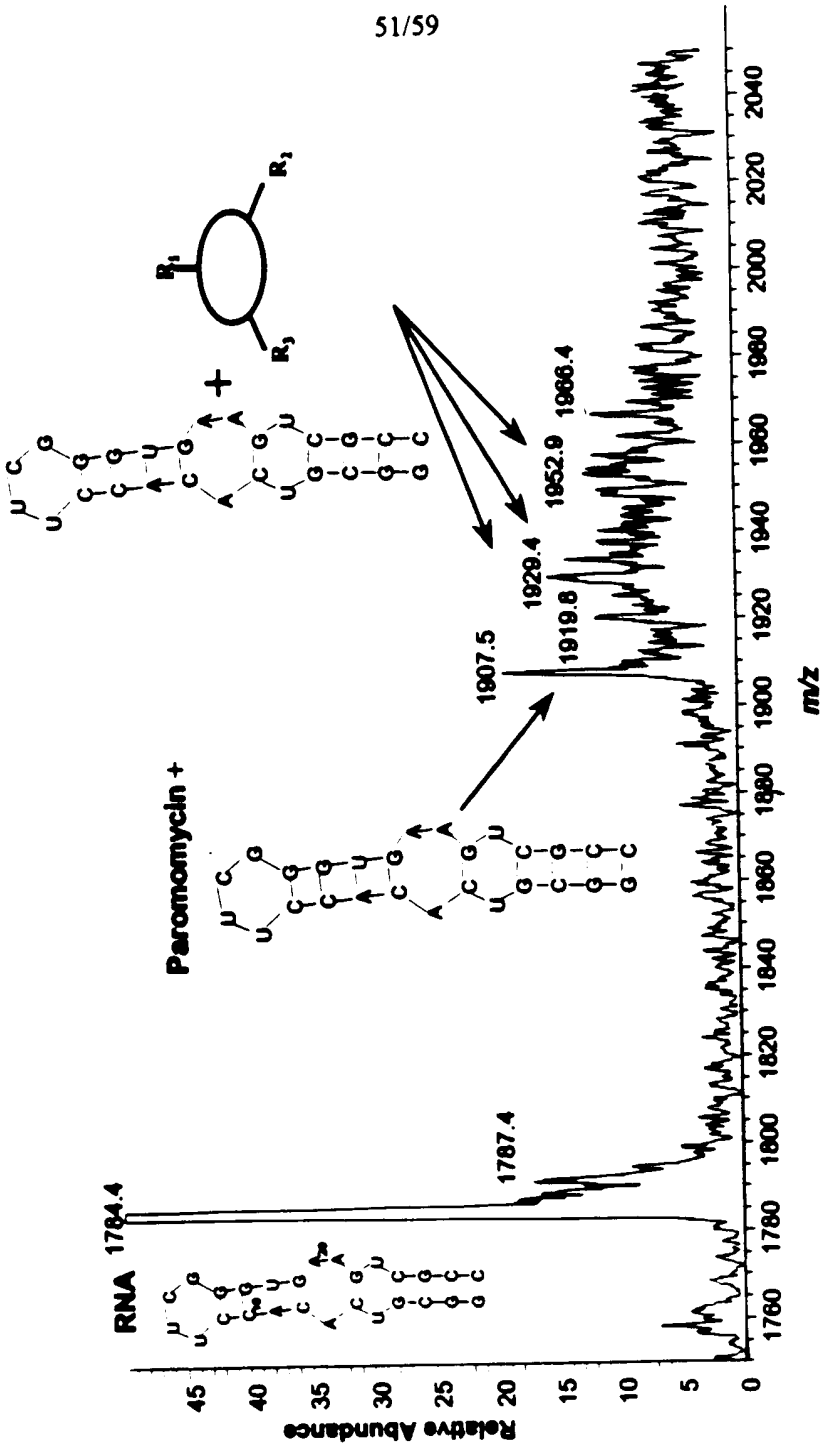

RNA
1784.4
1787.4
Paromomycin +
1907.5
1919.8
1929.4
1952.9
1966.4
Relative Abundance
m/z

Figure 44

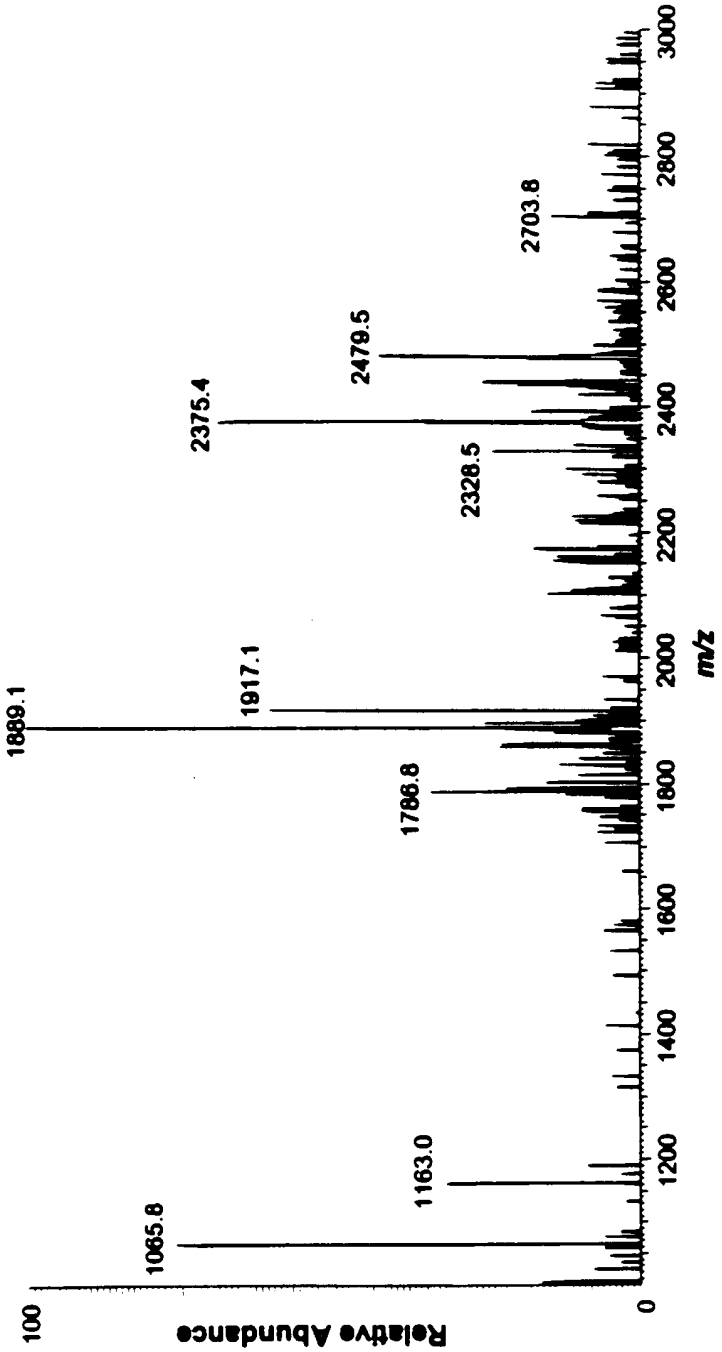

Relative Abundance
100
0
1065.8
1163.0
1786.8
1889.1
1917.1
2328.5
2375.4
2479.5
2703.8
1200
1400
1600
1800
2000
2200
2400
2600
2800
3000
m/z

Figure 45

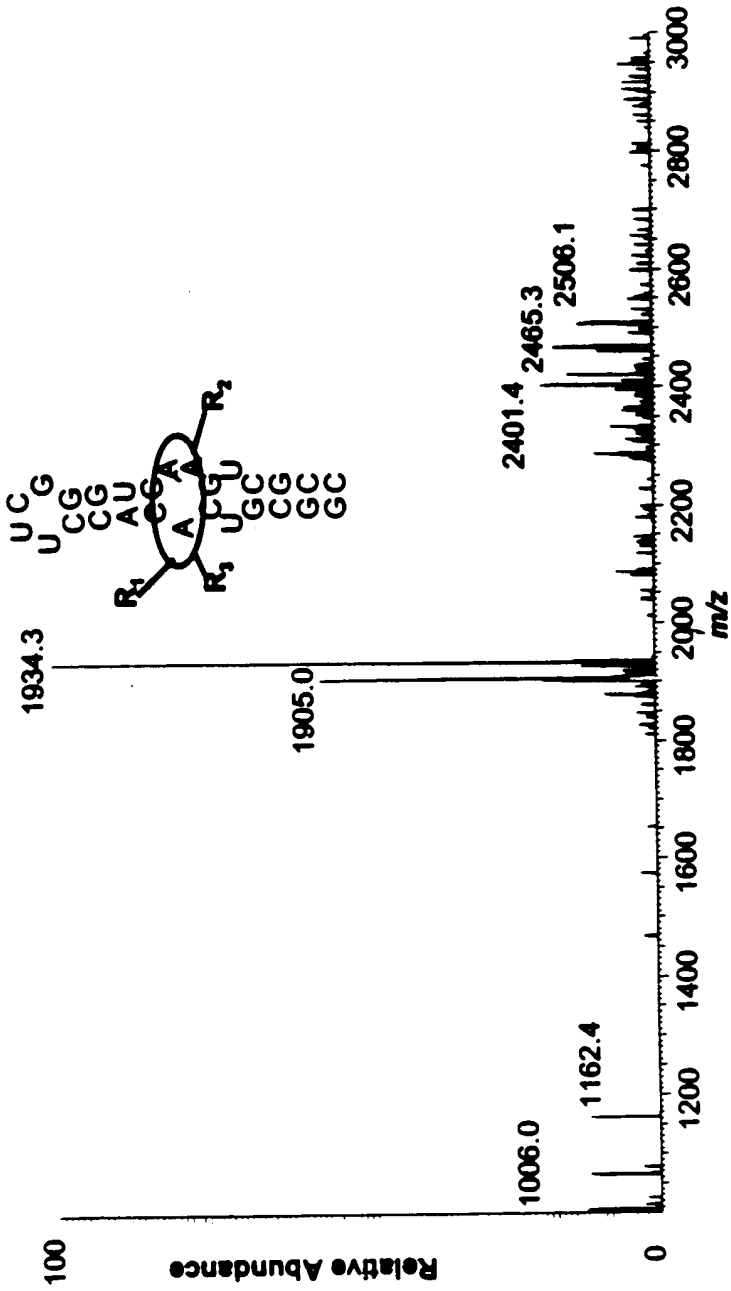

Relative Abundance
100
0
1006.0
1162.4
1905.0
1934.3
2401.4
2465.3
2506.1
1200
1400
1600
1800
2000
2200
2400
2600
2800
3000
m/z
R1
R2
R3

Figure 46